# Project Notes:

**Project Title: Parental Pharmacological Modulation of Serotonergic Signaling in C. elegans to analyze hereditary suicide.**
**Name: Charkam, Sahasra**

**Note Well:** There are NO SHORT-cuts to reading journal articles and taking notes from them. Comprehension is paramount. You will most likely need to read it several times, so set aside enough time in your schedule.

**Contents:**

# Knowledge Gaps:

This list provides a brief overview of the major knowledge gaps for this project, how they were resolved and where to find the information.

| Knowledge Gap | Resolved By | Information is located | Date resolved |
|---|---|---|---|
| Suicide Measurement | November 5th | Parallel serotonin between C. elegans and humans (helped decide assays for biological depression) | November 1st |
| Organisms of Use | November 8th | C. elegans article | November 8th |
| CRISPR-Cas9 | October 1st | CRISPR Article | September 29th |
| Serotonin Molecule pathways | October 1st | Molecular Basis of Suicide + Association of SERT polymorphism | September 18th |
| Assay use for experiment | November 30th | Youtube videos + articles that had the assays (in logbook) | Novemebr 28th |
| Use of C. elegans | November 18th | C. elegans article + additional resources (Dr. C) | November 15th |

| Knowledge Gap | How to Address |
|---|---|
| Suicide Measurement | Read the paper of the abstract I did for my Elevator Pitch - ISEF project |
| Organisms of Use | Research which organism is most feasible - given the time of 2 months, essentially look into the resources I have with Dr.C and look into what labs can provide me. |
| CRISPR-Cas9 | How does it work? Plan to find an expert in field (lab Principle investigator) and read articles |

| | |
|---|---|
| Serotonin Molecule pathways | Read the article - molecular basis of the suicidal brain (review article |
| *C. Elegans* | How do they work? How is their pathway similar to humans (serotonin)? Would they work as model organisms? |

# Literature Search Parameters:

These searches were performed between (Start Date of reading) and XX/XX/2019.
List of keywords and databases used during this project.

| Database/search engine | Keywords | Summary of search |
|---|---|---|
| PubMed | Suicide, Hereditary | Found the Hereditary basis of suicide – the first Article |
| PubMed | Suicide, Biology | Found the molecular, biological, and genetic basis of suicide. |
| Google Scholar | Suicide, Antidepressants, Serotonin | Found what medications work with and against Serotnin |
| Pubmed | Suicide gene, gene therapy, safety switch | Found research articles on suicide gene systems used to improve safety in gene and cell therapy |
| Google Scholar | CRISPR/Cas9, inducible cell death, cellular suicide switch | Identified studies describing inducible CRISPR-based suicide switches for controlled cell elimination – also found patents |
| Pubmed | Model organism, neuroscience, glial development | Found three model organisms to access which one would work best for my project |

# Tags:

| Tag Name | |
|---|---|
| Gene Networking | Biology |

| | |
|---|---|
| Suicide | Blood Brain Barrier |
| Depression/Anti-depressants | Genetics |
| CRISPR | SyntheticBiology |
| neuromodulation | invertebrate model |
| electrophysiology | nervous system |
| Aplysia Californica | neural interface, |
| CellularSafety | InducibleSwitch |
| GeneTherapy | |
| GutMicrobiota, | TryptophanMetabolism |
| AdolescentDepression | Neurotransmitters |
| Rna | Molecular Science |
| C.Elegans | Glial development |
| neurobiology | Epigenetics |
| DNAMethylation | Cognition |
| inflammation | antidepressants |
| cytokines | prognosis |
| primary care | neurochemistry |
| Adolescent | screening |
| CAR-T | SuicideGene |

| | |
|---|---|
| metabolism | AdoptiveCellTherapy |
| Neuropsychiatry | Depression |

# Article #0 Notes: Title

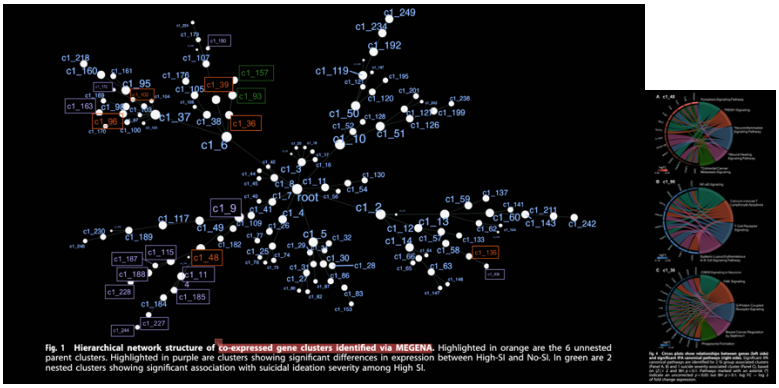
Article notes should be on separate sheets

**KEEP THIS BLANK AND USE AS A TEMPLATE**

| | |
|---|---|
| **Source Title** | |
| **Source citation (APA Format)** | |
| **Original URL** | |
| **Source type** | |
| **Keywords** | |
| **#Tags** | |
| **Summary of key points + notes (include methodology)** | |
| **Research Question/Problem/ Need** | |
| **Important Figures** | |
| **VOCAB: (w/definition)** | |
| **Cited references to follow up on** | |
| **Follow up Questions** | |

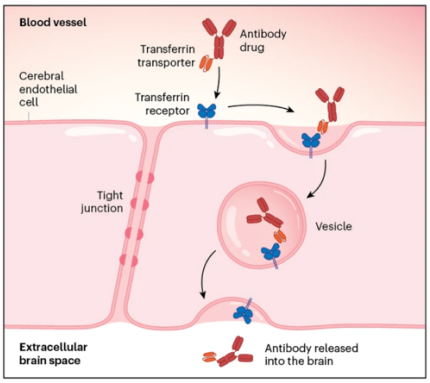
# Article #1 Notes: Use of Gene Networking to key in on specific RNA slices (Transcriptome) in correlation with suicide

| | |
|---|---|
| **Source Title** | **Brain and blood transcriptome profiles delineate common genetic pathways across suicidal ideation and suicide** |
| **Source citation (APA Format)** | Sun, S., Liu, Q., Wang, Z., Huang, Y., Sublette, M. E., Dwork, A. J., Rosoklija, G., Ge, Y., Galfalvy, H., Mann, J. J., & Haghighi, F. (2024). Brain and blood transcriptome profiles delineate common genetic pathways across suicidal ideation and suicide. *Molecular Psychiatry*, 1–10. https://doi.org/10.1038/s41380-024-02420-z |
| **Original URL** | https://www.nature.com/articles/s41380-024-02420-z |
| **Source type** | Primary Article |
| **Keywords** | Transcriptomes, blood-brain barrier, SI, gene co-expression, peripheral blood inflammation, grey and white matter |
| **#Tags** | Gene Networking, RNA |
| **Summary of key points + notes (include methodology)** | Given the hereditary characteristics of both suicidal ideation (SI) and behavior, the article focuses on the correlations between certain proteins and their impact on suicidal behaviors. The study collects data from 46 individuals in each of the three groups to use a gene network approach through gene co-expression patterns. Centralizing the bridge between SI and the specific RNA-sequence data in peripheral blood. Founding that 18 co-expressed modules are present in individuals who expressed SI, and an additional 3 co-expressed modules in SI severity showed researchers a strong association with the role of the brain and peripheral blood inflammation in suicide risk and heritability. The study uses two catagories of samples, split into living and postpartum. The living catagory is split into three groups: 1. A control group (No SI); 2. Individuals that have MDD without suicide ideations; 3. Individuals with MDD with SI. Postporetum individuals with suicide brains and naturally caused death brains are compared via their white and gray matter to isolate certain proteins which are linked with the suicide ideation. |
| **Research Question/Problem/ Need** | Correlation between key RNA transcriptomes and Suicide with a link to genetic heritability. |
| **Important Figures** | Fatemeh Haghighi |

| | |
|---|---|
| |  <br>Fig 1 is the hierarchial network structure of co-expressed gene clusters identified through MEGNA. Fig 4 are the circus plots that show the relationship between genes (left side) and significant IPA canonical pathways (right side). |
| **VOCAB: (w/definition)** | **Transcriptomes:** A part of RNA molecules expressed in organs, cells, tissues, etc.<br>**Suicidal Ideation:** The idea of committing suicide<br>**Abbrent gene:** a vital step in which there is tight regulation at steps such as transcription, translation, and splicing.<br>**Peripheral: Away from the center** |
| **Cited references to follow up on** | N/A |
| **Follow up Questions** | What types of gene editing techniques can be used to reverse/delete these specific proteins/transcriptors, such as the DNMT1 expression, to prevent suicide from being hereditary?<br><br>Are there any proteins known already that have been confirmed to cause either a high SI (Suicide Ideation) or attempts at self-harm?<br><br>How does the blood-brain barrier play a role in the permeation of chemicals (that would potentially negate these proteins? |

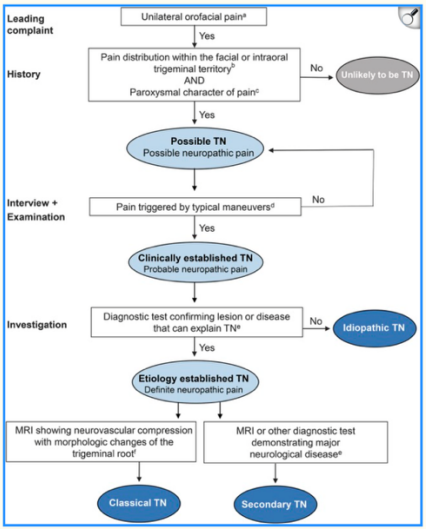
# Article #2 Notes: Brain drugs can now cross the once impenetrable blood–brain barrier (Summer Work)

| | |
|---|---|
| **Source Title** | Brain drugs can now cross the once impenetrable blood–brain barrier |
| **Source citation (APA Format)** | Abbott, A. (2025b). Brain drugs can now cross the once impenetrable blood–brain barrier. *Nature*, *641*(8065), 1086–1088. https://doi.org/10.1038/d41586-025-01569-z |
| **Original URL** | https://www.nature.com/articles/d41586-025-01569-z |
| **Source type** | Magazine Article |
| **Keywords** | Blood Brain Barrier, Brain shuttles, Transferrin Receptor, Therapeutic Delivery, Gene Therapy |
| **#Tags** | Blood Brain Barrier, Hunter's Syndrome |
| **Summary of key points + notes (include methodology)** | Hunter's Syndrome is a lysosomal storage disease caused by a sex-linked gene allele leading to the lack of an essential enzyme in the brain. The brain is protected by a tightly packed layer of endothelial cells lining the blood vessels, deriving is named the blood-brain barrier. The barrier only allows small fat fat-soluble, polar molecules through, preventing toxic molecules from entering. Traditional pharmaceutical approaches consist of creating small nonpolar molecules or utilizing existing transporters for smaller molecules. Less than 0.1% of the medication used for the brain penetrates the brain, leading to not only a waste of material but also very severe side effects, including the ones explained in the article for Hunter Syndrome (loss of hearing, thickening of facial features, loss of language, and more). The article explores a new approach to drug therapy and getting past the blood-brain barrier using brain shuttles. The most advanced brain shuttle involves the exploitation of the transferrin receptor system, which normally transports iron into the brain. These shuttles bind to receptors, enveloped in vesicles, travel across the cells, and then release the therapeutic molecule. The transferrin receptor system is precisely the area that the enzyme from Hunter's syndrome is missing, making this new drug therapy a plausible solution to surpassing the BBB. The 2021 enzyme replacement therapy for |

| | |
|---|---|
| | Hunter's Syndrome from JCR pharmaceuticals was the first ever and only authorized brain shuttle-based treatment, though not widely available much past Japan, which is what Denali Therapeutics plans on making available. This article relates to neuroscience and the permeability of the brain itself. The blood-brain barrier not only prevents molecules from going in but also stops molecules from going out, which sometimes might cause an issue. |
| **Research Question/Problem/ Need** | How can researchers safely and effectively deliver therapeutic molecules such as drugs, enzymes, or gene therapy across the blood-brain barrier. |
| **Important Figures** |  <br>The image shows an antibody drug that crosses the blood-brain barrier by binding to a receptor called transferrin on cerebral cells and are transported in vesicles and then released into the brain. |
| **VOCAB: (w/definition)** | Neuropharmacology:<br>Cerebrospinal Fluid:<br>Amyloid Proteins: |
| **Cited references to follow up on** | N/A |
| **Follow up Questions** | How can these genes that are transported to the blood brain barrier be used in suicide prevention. |

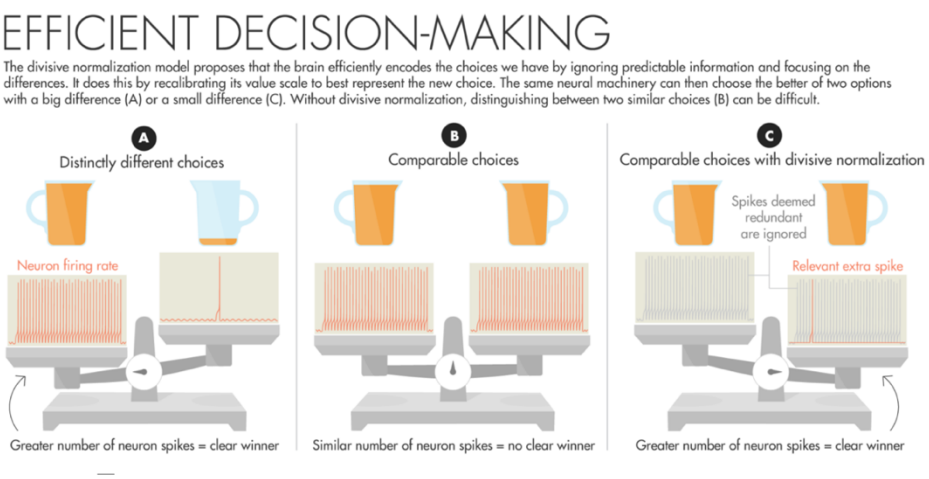
# Article #3 Notes: Trigeminal neuralgia New classification and diagnostic grading for practice and research (Summer Work)

| | |
|---|---|
| **Source Title** | Trigeminal neuralgia New classification and diagnostic grading for practice and research |
| **Source citation (APA Format)** | Cruccu, G., Finnerup, N. B., Jensen, T. S., Scholz, J., Sindou, M., Svensson, P., Treede, R.-D., Zakrzewska, J. M., & Nurmikko, T. (2016). Trigeminal neuralgia. *Neurology*, *87*(2), 220–228. https://doi.org/10.1212/wnl.0000000000002840 |
| **Original URL** | https://www.neurology.org/doi/pdfdirect/10.1212/WNL.0000000000002840 |
| **Source type** | Review Article |
| **Keywords** | Neuropathic, Nosology, Etiology, Paroxysm, Idiopathic |
| **#Tags** | Trigeminal Neuralgia |
| **Summary of key points + notes (include methodology)** | Trigeminal Neuralgia (TN) is a neurological disorder of the trigeminal nerve (which is present under the brainstem), causing neuropathic facial pain. Classification of TN has taken an increasing toll on the medical community due to inconsistent terminology and its unalignment with the current grading scale for neuropathic pain and headache disorders. The article focuses on three proposed classifications of TN: Classical TN, diagnosed by evidence of the morphological change in the trigeminal nerve root caused by vascular compression; Secondary TN, diagnosed by a known underlying neurological condition, like MS or a tumor; and finally Idiopathic TN, diagnosed with unknown cause (etiology) after extensive investigation. A possible grading system was also introduced: Possible TN, with minimal characteristics including pain remaining unilateral (unless in the case of TN with MS) and distributed through facial or intraoral trigeminal territory (not to extend towards cervical nerve area); Clinically Established TN, characterized by the pain paroxysms triggered by mechanical stimuli or facial movement; and finally Etiology Established TN, known as definite neuropathic pain, is the highest level of diagnostic certainty in which imaging/diagnostic testing (usually MRI) confirms and underlying neuropathic issue that results in the diagnostic of classical or secondary TN. This article pertains to my ideas because it allows me to think of the types of categories when analyzing an |

| | |
|---|---|
| | issue. Beyond that, TN is also known to destroy myelin sheath within the brain, connecting back to my idea of the linkage of myelin sheath in MS and how TN is connected to both MS and the destruction of myelin sheath. |
| **Research Question/Problem/ Need** | The article aimed to find new classifications and diagnostic grading for Trigeminal Neuralgia. |
| **Important Figures** |  <br>The image outlines the process in which TN is diagnosed in a very concise manner. |
| **VOCAB: (w/definition)** | Paroxysms: A sudden attack or recurrence of symptoms, often referring to pain or convulsions<br>Neuropathic: Relating to damage or dysfunction of nerves that causes pain<br>Etiology: The cause or origin of a disease |
| **Cited references to follow up on** | N/A |
| **Follow up Questions** | How can these category breakdowns relate to suicide and different aspects of suicide. |

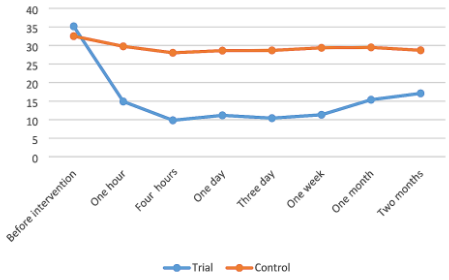
# Article #4 Notes: The Neuroscience Behind Bad Decisions (Summer Work)

| | |
|---|---|
| **Source Title** | The Neuroscience Behind Bad Decisions |
| **Source citation (APA Format)** | Singer, E. (2016, August 23). *The Neuroscience Behind Bad Decisions*. Quanta Magazine. https://www.quantamagazine.org/the-neuroscience-behind-bad-decisions-20160823/ |
| **Original URL** | https://www.quantamagazine.org/the-neuroscience-behind-bad-decisions-20160823/ |
| **Source type** | Magazine Article |
| **Keywords** | Neuroeconomics, Value Encoding, Energy Efficencency, Heuristics, Framing effects |
| **#Tags** | Neuroeconomics |
| **Summary of key points + notes (include methodology)** | The science of irrational decision making is hypothesised to be linked with the brain's substantial energy requirements. Traditionally economists believed that humans assign values to items and tend to pick the item with a higher score. But, neuroscientists suggest that the brian holds the answer to this decision making. Gilmer's Theory, a theory based on neuroscience's errors in decision making. It argues that the brain prioritizes efficiency over decision making in order to optimize the number of neuron firings or "spikes" that occur. The model is based on the "efficient coding hypothesis" which when applied, Gilmer proposes, the brain recaliberates its scale in order to make clear distinctions between two similar choices. This is known as divisive normalization which implies that neurons encode only the relative differences between choices, effectively taking out redundant information to conserve energy. This new field of neuroeconomics is filled with ongoing controversies, such as Dr. Yu believes the brain works on a more intricate level than that of the visual system, making it more complex than simply information gathering. This article allows me to discover new subtopics that can be paired with neuroscience and the various fields that are associated with it. The field of neuroeconomic is relatively new, allowing me more wiggle room in a question to research that is unique and innovative. |

| | |
|---|---|
| **Research Question/Problem/ Need** | What do humans often make suboptimal decisions and how do brain processes, neural computations, and energy constraints contribute to these bad decisions. |
| **Important Figures** |  <br>The image shows brain uses divisive normalization to make efficient decisions by filtering out redundant information, to distinguish between similar choices more clearly. |
| **VOCAB: (w/definition)** | Neuroeconomics: Interdisciplinary field combining neuroscience and economics to understand decision-making processes<br>Value Encoding: The brain's method of assigning values to choices which can be influenced by context and presentation<br>Energy Efficiency: The brain's preference for energy-saving strategies, sometimes at the cost of optimal decision making<br>Heuristics: Mental shortcuts that simplify decision-making but can lead to biases and errors. |
| **Cited references to follow up on** | N/A |
| **Follow up Questions** | How can neuroeconomics impact the decisions that induvials with mood disorders make. |

# Article #5 Notes: Rapid and sustained antidepressant effects of intravenous ketamine in treatment-resistant major depressive disorder and suicidal ideation: a randomized clinical trial

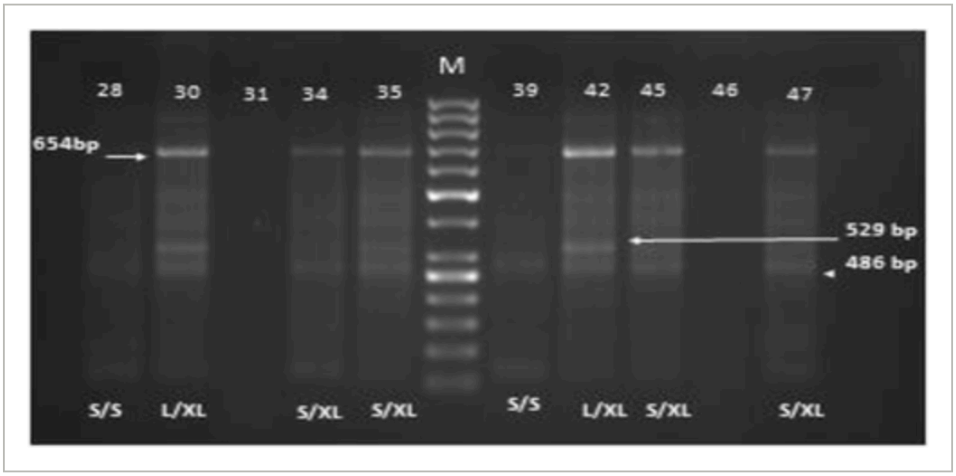
| | |
|---|---|
| **Source Title** | Rapid and sustained antidepressant effects of intravenous ketamine in treatmentresistant major depressive disorder and suicidal ideation: a randomized clinical trial |
| **Source citation (APA Format)** | Zolghadriha, A., Anjomshoaa, A., Jamshidi, M. R., & Taherkhani, F. (2024). Rapid and sustained antidepressant effects of intravenous ketamine in treatment-resistant major depressive disorder and suicidal ideation: a randomized clinical trial: BMC Psychiatry. *BMC Psychiatry*, *24*(1), 1–8. https://doi.org/10.1186/s12888-024-05716-0 |
| **Original URL** | https://bmcpsychiatry.biomedcentral.com/articles/10.1186/s12888-024-05716-0 |
| **Source type** | Primary Research Article |
| **Keywords** | Ketamine, Major depressive disorder, Suicide ideation, Clinical trial |
| **#Tags** | Ketamine, MDD, Suicide Ideation |
| **Summary of key points + notes (include methodology)** | Major Depressive Disorder (MDD) is a highly disabling issue with significant economic and personal burdens. Around one third of patients are treatment-resistant and many medications take weeks to show effect. Ketamine has risen as a rapid acting antidepressant with potential anti-suicidal effects. The study was a randomized, controlled, assessor-blinded clinical trial. It was conducted in Iran (from April to August 2022) with 64 patients who were diagnosed with treatment resistant MDD. Participants were randomly assigned two groups, Ketamine group (0.5mg/kg IV infusion) and Placebo group (normal saline). Both groups continued their anti-depressants. The researchers used the Montgomery-Asberg Depression Rating Scale (MADRS) scale to measure depression, The Beck's Suicidal Ideation Scale (BSIS) for suicidal Ideation, and followed up with assessments every 1hr, 4 hrs, 1 day, 3 days, 1 week, 1 month, and 2 months post infusion. The results showed rapid and significant improvement of |

| | |
|---|---|
| | Deepression within one hour of the ketamine infusion and was also the same 2 months after (with a p value less than 0.001). Simultenously there was also a reduction in suicidal ideation in the ketemaine group post infustion with a p value of less than 0.001 making it significant. However there were side effects which included headaches/dissociative symptoms, dizziness, nausea, anxiety, and visual disturbances. The study confirmed that rapid onset and sustained antidepressant/antisuicidal effects of ketamine. Furthermore it suggests the mechanisms of katamine itself in which it may enhance neroplasticty, glutamate modulation, anti-inflammatory effects. In conlusion a single IV dose of ketamine provides rapid relif of depression and suicide ideation for weeks and months on end. Furhter research would be needed on long-term safety, how to dose the meducationa dn predictors of treatment responses. |
| **Research Question/Problem/ Need** | To investigate the clinical effect of intravenous ketamine on symptoms of MDD and suicidal ideation. |
| **Important Figures** |  <br>**Fig. 2** The trend of depression score in trial (ketamin), and control (placebo) groups<br><br>**Table 4** Comparison of suicidal ideation score before, and after treatment<br><br>\| \| Trial \| Controls \| *p*-value \|<br>\|---\|---\|---\|---\|<br>\| Before intervention \| 6.74 ± 6.67 \| 3.58 ± 2.57 \| 0.11 \|<br>\| After intervention \| 0.42 ± 1.52 \| 3.35 ± 2.80 \| < 0.001 \|<br><br>Figure 2 shows the trend of depression scores in the ketamine trial and the control group. Table 4 shows comparison of suicidal ideation score before and after treatment. |
| **VOCAB: (w/definition)** | Allocation: The action/ process of disturbing something<br>Placebo: A group that is used as a control, so it is not a true treatment group but usually administered with saline or water or a drug that has no effect at all.<br>Intravenous: Existing or taking place within |
| **Cited references to follow up on** | Nikkheslat N. Targeting inflammation in depression: ketamine as an anti-inflammatory antidepressant in psychiatric emergency. Brain Behav Immunity-Health. 2021;18:100383. |
| **Follow up Questions** | Does Ketamine have similar function to ibuprofen in the sense of anti-inflammation? |

# Article #6 Notes: Association of serotonin transporter gene polymorphism with efficacy of the antidepressant drugs sertraline and mirtazapine in newly diagnosed patients with major depressive disorders

| | |
|---|---|
| **Source Title** | Association of serotonin transporter gene polymorphism with efficacy of the antidepressant drugs sertraline and mirtazapine in newly diagnosed patients with major depressive disorders |
| **Source citation (APA Format)** | Gulfishan, S., Halder, S., Kar, R., Srivastava, S., & Gupta, R. (2022). Association of serotonin transporter gene polymorphism with efficacy of the antidepressant drugs sertraline and mirtazapine in newly diagnosed patients with major depressive disorders. *Human Psychopharmacology: Clinical and Experimental*. https://doi.org/10.1002/hup.2833 |
| **Original URL** | https://pubmed.ncbi.nlm.nih.gov/35089613/ |
| **Source type** | Primary Research Article |
| **Keywords** | BDI score, MDD (major depressive disorder), SERT gene polymorphism |
| **#Tags** | Depression, Serotonin, Suicide |
| **Summary of key points + notes (include methodology)** | Manic Depressive Disorder (MDD) is one of the lead causes of burden among all disease - only surpassed by lower respiratory infection and HIV/AIDS. MDD causes disability and is associated with high mortality and morbidity. The cause of depression varies from psychological, genetic, or biochemical. Hypothesis 1: The monoamine hypothesis supposes that deficiency of monoamines (serotonin) is a cause of depression - deficit in monoamine function can be restored by antidepressant drugs such as selective serotonin re-uptake inhibitors (SSRIs). Hypothesis 2: Hypothesizes the role of neurotropic and endocrine factors in pathophysiology of |

| | |
|---|---|
| | depression. Evidence suggests a decreased central serotonergic turnover is associated with MDD. The reduced level of primary serotonin metabolite 5-hydroxy indoleacetic acid (5-HHTLPR) in the cerebrospinal fluid has been associated with violent and impulsive behavior (suicidal tendencies). Many patients have poor response and adverse effects to antidepressants. Recent research shows involvement of Serotonin receptor Transporter (SERT) gene neurotransmitter transporter in regulating the efficacy of antidepressant therapy. SERT is part of the SLC6A4 variant like 5-HTTLPR, and individuals with a homozygous small allele are more susceptive to MDD than those with the long gene. Blocking the serotonin transporter is the main mechanism of SSRI's, hence the gene encoding this protein is a strong area of research. Several reports suggest that 5HTTLPR polymorphism plays a role in depression, the s allele is believed to exercise both direct and indirect effect on the severity of depression, short variant is associated with poorer response to antidepressant treatments. However contradictory findings in Korean and Japanese populations found that the short allele is associated with favorable outcomes. Two antidepressant drugs chosen were Sertraline (ST) - inhibits reuptake of 5-HT in CNS with few adverse effects - and Mirtazapine (MZ) - noradrenergic and specific serotonergic antidepressant with higher remission, faster onset, less side effects, also effective in augmentation therapy. They hypothesized Selected MZ and ST to analyze association of SERT gene polymorphism and clinical response in newly diagnosed, treatment naive MDD patients in the northern region of Indian subcontinent. The Study Protocol was approved by the Institution Ethical Clearance Committee, and written informed consent was obtained. This was a randomized, open labeled prospective observational study. Inclusion Criteria: DSM-5 first episode of MDD, BDI ≥21. Exclusion: acute suicidal risk, DSM-5 dementia, schizophrenia, schizoaffective or bipolar diseases, PTSD, OCD, anxiety, eating disorder, antidepressant use in 3 months, substance dependency, organic brain disease, pregnancy, lactation, significant medical illness. 80 patients randomized (40 MZ, 40 ST). Doses: MZ (14–45 mg/day), ST (25–200 mg/$_{day}$). BDI assessed baseline and 6 weeks. Positive response = ≥50% BDI reduction. DNA extracted, quantified, PCR for 5-HTTLPR with primers (Forward 5'-GGC GTT GCC GCT CTG AAT GCC A-3', Reverse 5'-GAG GGA CTG AGC TGG ACA ACC AC-3'). PCR products separated by gel electrophoresis: Short (S; 486 bp), Long (L; 529 bp), Extralong (XL; 612/654 bp). Genotypes: S/S, S/L, L/L, S/XL, L/XL. Statistical analysis by ANOVA, Chi-square/Fisher's exact, SPSS v20, $p<0.005$ significant. Results: MZ baseline BDI 27.30 ± 3.67 reduced to 17.07 ± 3.30 ($p<0.057$). ST baseline 28.48 ± 3.38 reduced to 18.35 ± 3.50 ($p<0.057$). Both groups showed significant reduction at 6 weeks ($p<0.001$). Response rates: ST 88%, MZ 95%. MZ best in L/L, XL/XL, poorer in S/S, S/L, XL/L. ST best in heterozygous (S/L, XL/L, XL/XL), least in L/L and S/S. No significant difference between drug groups by genotype ($p=0.611$). 26/80 patients (32.5%) were S/S, showing higher MDD risk. Compared to XL/XL, S/S showed greater prevalence and less BDI reduction, consistent |

| | |
|---|---|
| | with evidence linking S/S to higher depression risk and poor treatment response after stress. Other studies also showed S/S increases MDD risk compared to L/L. In conclusion, drug response in MDD depends on genotype, with MZ better in L/L and XL/XL and ST in heterozygotes, while S/S was linked to poor response and higher susceptibility. Findings can help clinicians predict antidepressant responsiveness and recommend SERT-targeted treatment for better therapeutic outcomes. |
| **Research Question/Problem/ Need** | To find the association of serotonin receptor transporter gene polymorphism in patients with MDD with the clinical delicacy of Mirtazapine (MZ) and Sertraline (ST). |
| **Important Figures** |  <br>Group 1<br>Sertaline<br>Genotypes \| 0 (S/S) *n* = 13 \| 1 (S/L) *n* = 5 \| 2 (L/L,S/XL) *n* = 9 \| 3 (L/XL) *n* = 9 \| 4 (XL/XL) *n* = 4<br>BDI difference \| 9.61 ± 3.30 \| 11.80 ± 3.89 \| 7.55 ± 3.28 \| 11.77 ± 2.90 \| 11.75 ± 2.87<br>Group 2<br>Mirtazapine<br>0 (S/S) *n* = 13 \| 1 (S/L) *n* = 3 \| 2 (L/L, S/XL) *n* = 11 \| 3 (L/XL) *n* = 12 \| 4 (XL/XL) *n* = 1 \| *p* value (within groups) \| *p* value (between groups)<br>8.84 ± 3.15 \| 9.33 ± 2.30 \| 12.90 ± 4.67 \| 9.33 ± 5.06 \| 12.00 ± 00 \| 0.644(NS) \| 0.989(NS)<br>Figure 3 shows Agrose gel electrophoresis images of DNA products separated by alleles.<br>Table 1 shows the genotype versus the BDI scores between the two medications. |
| **VOCAB: (w/definition)** | BDI – Beck Depression Inventory Score is a widely used self-report questionary with a total of 21 questions, 3 points each, with higher scores |

| | |
|---|---|
| | indicating more severe depression.<br>SERT Gene Polymorphism – Variations of Serotonin transporter gene and how it affects serotonin signal molecule in brian. Certain variants have been linked to increased susceptibility to depression and difference in response to antidepressants.<br>Geneotypic Expression – Observable pattern of gene's variants (genotype) and how they manifest in an individual. This aricle refers to which version of the SERT gene is expressed. |
| **Cited references to follow up on** | Margoob, M. A., & Mushtaq, D. (2011). Serotonin transporter gene polymorphism and psychiatric disorders: Is there a link? Indian Journal of Psychiatry, 53(4), 289–299. |
| **Follow up Questions** | How can this medication be different to that of gene therapy and directly increasing serotonin signaling molecules? |

# Article #7 Notes: Genetic Contributions to Suicidal Thoughts and Behaviors

| | |
|---|---|
| **Source Title** | Genetic Contributions to Suicidal Thoughts and Behaviors |
| **Source citation (APA Format)** | DiBlasi, E., Kang, J., & Docherty, A. R. (2021). Genetic contributions to suicidal thoughts and behaviors. *Psychological Medicine*, *51*(13), 1–8. https://doi.org/10.1017/s0033291721001720 |
| **Original URL** | https://pmc.ncbi.nlm.nih.gov/articles/PMC8477225/#:~:text=Familial%20research%20has%20indicated%20that,Voracek%20%26%20Loibl%2C%202007). |
| **Source type** | Review Article |
| **Keywords** | Suicide, suicide attempt, suicidal ideation, suicidal thoughts and behaviors, genetics, genetic epidemiology |
| **#Tags** | Suicide, Biology, Genetics |
| **Summary of key points + notes (include methodology)** | Suicide is a global issue, impacting over half the world, it has been an increasing issue, with the US loosing over $70 billion dollars in annual medical expenses and losses. Suicide is complex and multidimensional involving physical, environmental, psychiatric, and genetic factors. Additionally suicide is not in it's own category but rather a part of major depression/borderline personality disorder which makees measuring suicide increasingly hard. There were three types measures: Suicide Ideation (SI), Sucide Attempt (SA) and Suicide Death. SI is the thoughts of ending one's life, whereas SA is non-fatal self harm with intent to dies. Suicide death is fatal injury that leads to death. The purpose of the paper is to review eveidnce that STBs (Suicidal thoughts and behavior) are heritable trats. Beyond that they also summarizre findings on family gentics and how suicide is passed down. They did this through 1. Epidemiological and Biometrical Studies. This involves family studies and figuring out how genetics with in fmailies are relations. Specifixally they focused on registry studies in Swedish population. Further 2. Molecular Genetics through Genome-wide Association Studies (GWAS) with common gentics variants (SNPs Single Nucleotide Polymorphism) and also conducted large international consortia (genome consortium). 3. They took whole-exome sequencing and CNVs (Copy number variants) to examine differences |

| | |
|---|---|
| | between different genomes between patients to find rare variants. In the family data they found STB heritibility estimates between 30-55%, and indivuals with suicide that runs in the family can be passed on and the chid is more likely to experience suicide. The swedish population showed genetics as the primary dirver suicide intergenerational transmission. Furthermore through GWAS for suicide ideation they found three genome-wide significatn loci Chr9: rs62535711, Chr11: rs598046, Chr13: rs7989250 within the chromosomes, and a SNP-based hertibility of 7.6%. For Suicide attempts (psyciatric genomic consortium) they found 3 loci - Chr10: rs45593736 (MDD cohort), Chr4: 23273116_D (BPD cohort), Chr2: rs138689899 (MDD+BPD). Genetic risk over lap between MDD and suicide attempts. Finally for suicide death they noticed 2 loci - Chr13: rs34399104, Chr15: rs35256367 with an SNP heritbility of 25%. Rare protien coding variants (>2,600 suicide deaths) implicated genes in cell cycle control and DNA repair. Further more CNVs linked suicide attemps to smaller studies including those with mood disorder loci. The most common pathwyas in which had 2500 genes associated in suicide behavior overall include gluscose egulation, protient localization, and cell cycle/DNA repair. Overall there are polygentic natures of suicide that cause ut to be heritable, with many different variaants of genes that paly key roles in suicide. The key candiates genes in suciide research are seotonin systems (SLC6A4 → serotonin transporter gene – leading to mood, impulsivity and aggrestion), Dopamine system ( DRD2 → dopmaine receptor D2 – reward, motivation, impulsivity), neutropic and plasitic genes ( BDNF – brain derived nerotropic factors), and Stress response (HPA axis gene – FKBP5 – which regulwates glucocoticoid receptor sensitivity). In conclusion this article highlisghts the specific pathwya and genes that are a part of polygentic suicide in hereditary sucide ideation, attempts, and death. |
| **Research Question/Problem/ Need** | The review article focuses on the hereditary asspect of suicide and the specific chromosomes linked to suicide. |
| **Important Figures** | Emily Diblasi<br>**Table 1.** Overview of GWAS efforts with genome-wide significant findings in STBs<br>Table 1 shows Suicidal ideation, suicidal attempts, chromosomes, and major depressive disorder, and bipolar disorder, with an overview of GWAS efforts with genome wide significant findings in STBs. |

| Phenotype | Ascertainment | Samples | Genome-wide significant loci number, chromosome location and SNP identifier | Study |
|---|---|---|---|---|
| Suicidal ideation, self-harm and suicide attempt | Population-based, European ancestry | • 36 599 SI<br>• 2498 self-harm; unknown suicidal intent<br>• 2666 SA<br>• 83 557 controls | 3 Total<br>Chr9:rs62535711; Chr11:rs598046; Chr13: rs7989250 | Strawbridge et al. (2019) |
| Suicide attempt | Clinical cohorts with diagnoses of major depression, bipolar disorder, or schizophrenia, European ancestry | Major depressive disorder<br>• 1622 SA<br>• 8786 controls<br>Bipolar disorder<br>• 3264 SA<br>• 5500 controls<br>Schizophrenia<br>• 1683 SA<br>• 2946 controls | 3 Total<br>MDD: 1<br>Chr10:rs45593736<br>BPD: 1<br>Chr4:23273116_D<br>MDD + BPD:1<br>Chr2:rs138689899 | Mullins et al. (2019) |
| Suicide death | Population-based, European ancestry | • 3413 suicide deaths<br>• 11 049 controls | 2 Total<br>Chr13:rs34399104; Chr15: rs35256367 | Docherty et al. (2020) |

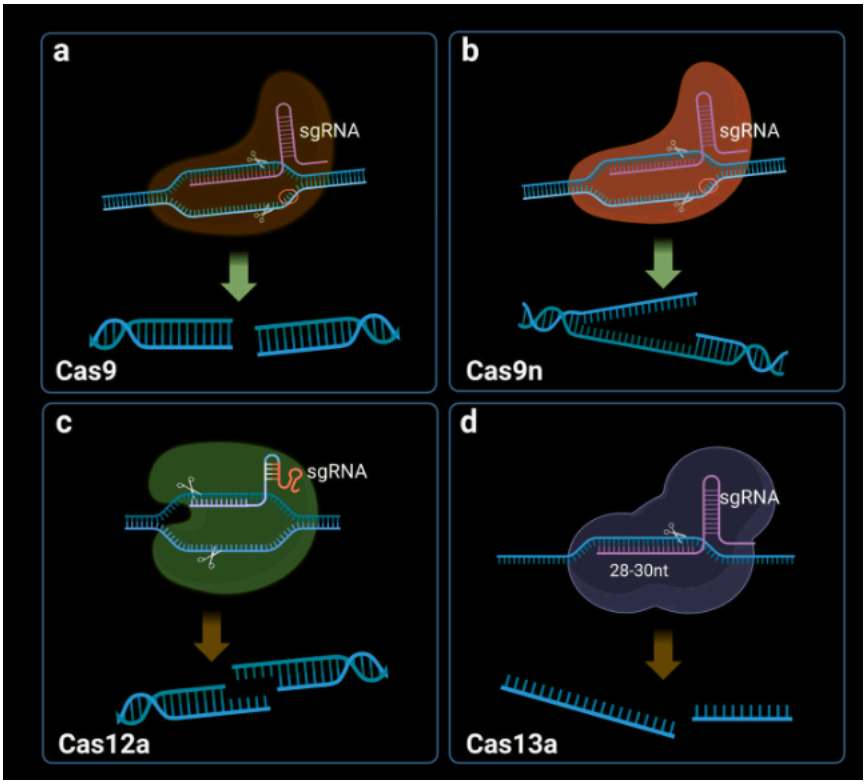
SI, suicidal ideation; SA, suicide attempt; Chr, chromosome; MDD, major depressive disorder; BPD, bipolar disorder.

| | |
|---|---|
| **VOCAB: (w/definition)** | Epidemiology: Study of incidence, distribution and possible control of diseases and other factors related to health<br>Serotonin: A neurotransmitter and hormone that plays a vital role in regulating mood, sleep, learning, appetite, and memory |
| **Cited references to follow up on** | 1. Brent, D. A., Melhem, N. M., Oquendo, M., Burke, A., Birmaher, B., Stanley, B., … Mann, J. J. (2015). Familial pathways to early-onset suicide attempt: A 5.6-year prospective study. JAMA Psychiatry, 72(2), 160–168. 10.1001/jamapsychiatry.2014.2141. [DOI] [PMC free article] [PubMed] [Google Scholar] |
| **Follow up Questions** | How can I use chromosomal pass downs in my project to test suicide hereditary risk. |

# Article #8 Notes: CRISPR/Cas9 Therapeutics: Progress and Prospect

| | |
|---|---|
| **Source Title** | CRISPR/Cas9 Therapeutics: Progress and Prospect |
| **Source citation (APA Format)** | Li, T., Yang, Y., Qi, H., Cui, W., Zhang, L., Fu, X., He, X., Liu, M., Li, P., & Yu, T. (2023). CRISPR/Cas9 therapeutics: Progress and Prospects. Signal Transduction and Targeted Therapy, 8(1), 1–23. https://doi.org/10.1038/s41392-023-01309-7 |
| **Original URL** | https://www.nature.com/articles/s41392-023-01309-7 |
| **Source type** | Review Article |
| **Keywords** | CRISPR, Cas9, PAM, Off-targeted Effects, Prime Editing, Delivery Vectors |
| **#Tags** | CRISPR (Clustered Regularly Interspaced Short palindromic Repeats) |
| **Summary of key points + notes (include methodology)** | Genome editing is currecntly revolutionzing medicine as it lets researchers directly alter DNA inside living cells. CRISPR/Cas9 stands out for the simplicity, versitiliaty and the cost compared to other genetic variation tools such as Zinc-Finger nucleases and TALENs. CRISPR uses a RNA to guide the nuclease, targeting any gene. CRISPR began in 1987 with repetitive DNA sequences (later called CRISPR – in E.Coli). Then CRISPR spacers began to be found to match viral DNA (to prove the similarities between CRISPR and bacterial immune systems). Then in 2013 CRISPR/Cas9 was first used in mammalian cells which led to a cascade of experiments to be engineered and new variants of the CRISPR system inclusing Cas12 and Cas13. The first human trial was conducted in CHINA in 2016 with the editing of T-cells targeting PD-1. There are three main parts of the CRISPR system. The main protein that cuts DNA or RNA is the called the Cas Nuclease – this is known as the Cas9, (streptococcus pyogenes version – spCas9). The Cas 9 recognizes a specific DNA sequence called the PAM (prostospacer adjacent motif – any base followed by two “GG”) – essentially makes sure that random cuts are not made. The Cas9 has two main nuclease domains (RuvC and HNH) which work together to create a double-strand break, with lots of variants that exist. The second part is the guide RNA (gRNA or sgRNA) which is a short synthetic RNA (around 20 nucleotides) that directs Cas9 to target DNA by complementary base pairings. In bacteria the system uses two RNAs (crTNA and tracrRNA) – in lab called sgRNA for simplicity. The gRNA (guide RNA) provides the programable target function which changes the RNA sequence in the target site. The third part is the Repair template which occurs when the Cas9 makes a break the cell repairs it through two ways: one is the non-homologugus end joining which leads to fast but errors often |

| | |
|---|---|
| | creating insertions or deletions that disrupt the gene, or two which is homology-direct repair (HDR) which uses a pre-exsisting DNA template to precisely insert/correct a sequence (less efficent in mammalian cells).<br>There are variants in Cas9 itself due to its limitatiosn which include the fact that Cas9 can only recognize sites with NGG PAM, cuts uninteded sites, and is very large and hard to package into small viral vectors like AAV. Hence there are a few variants. Cas9 nickase is moleucle with one disabled doamin which means it only cuts on strand, which allows this approach to reduce off-target cutting. Another one is dead Cas9 which happens when both nuclease domains are disabled so it can still bind to DNA but can not cut which is cruical for gene regulation studies and is being explored as a therapy for diseases where turning genes up or down is more useful than editing them. A third ariation is the Cas12a which has a different class neucleas, it recognizes T-rich PAMs which expands possible target sites, cuts DNA in a staggered way, and only requires a small amount of CRISPR RNA (crRNA) and no tracrRNA. Another variation is the Cas13 which is an RNA targeting enzyme which focuses on RNA transcripts. Further more there are many variaatiosn in Cas9 to make it more approachable for different asspects of gene editing. There are also base editors which edit specific bases and prime editing which finds and replaces base substiutions using Cas9 nickase (fused with reverse transcriptase) and a prime editing guide RNA. Delivery is increasingly important because the DNA needs to survive in bloodstreams without being degraded, reach the correct tissue/enter the correct cell, and cross the cell membranes and make it into the DNA nucleus., and do all this without harming the body. Some examples of drug delivery include Viral Vectors (such as AAVs), Lipid Nanoparticles (LNPs like lipids/cholestrol), Polyethelenimine (PEI such as a cationic polymer), Exosomes (such as tiny membrane bound vescicles that are natrually secreated by cell), and Biomimetic nanoparticles (which hide in natural cell membranes improve targets). There are theraputic applications in the Liver, blood, cancer and infectious dieases (which is where Cas13 would be used). There have also been various clinical trials including blood stem cells and T cells. Overall CRISPR has been from bacterial immunity to potential human genetic therapy in such a short time, with early clinical trials showing major advances in gene editing therapeutics. |
| **Research Question/Problem/ Need** | The article reviews how CRISPR/Cas9 gene editing technology is being developed and used to treated diseases, and look at the progress that has been done so far. |
| **Important Figures** | Tianxing Li (Author) |




Schematic diagram of DNA strand cleavage tools.

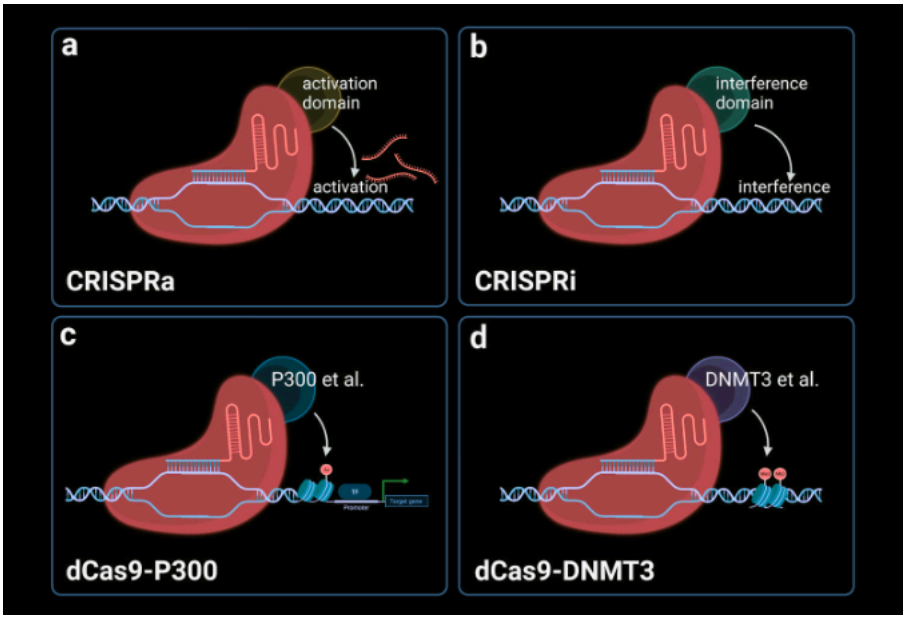



Diagram of dCas9-based tools to regulate expression (need to use something similar to regulate serotonin protien)

| | |
|---|---|
| **VOCAB: (w/definition)** | Delivery Vectors: Systems used to get CRISPR into cells (viruses, lipid nanoparticles, exosomes)<br>Off-Targeted Effects: Unintended DNA cuts or edits at sites similar to targer |

| | |
|---|---|
| | (huge safety concern)<br>AAV Vectors: Adeno-associated Virus small harmless viruses that commonly used as delivery vehicles in gene therapy.<br>Duchenne muscular dystrophy: Severe genetic disorder caused by mutations in the dystrophin gene which is essential for muscle strength and stability. |
| **Cited references to follow up on** | N/A |
| **Follow up Questions** | How can CRISPR be used to up or down regulate specific moleucles in the brain to increase/decrease suicide. |

# Article #9 Notes: Biological Basis of Suicide and Suicidal Behavior

| | |
|---|---|
| **Source Title** | Biological Basis of Suicide and Suicidal Behavior |
| **Source citation (APA Format)** | Pandey, G. N. (2013). Biological basis of suicide and suicidal behavior. *Bipolar Disorders*, *15*(5), 524–541. https://doi.org/10.1111/bdi.12089 |
| **Original URL** | https://onlinelibrary.wiley.com/doi/10.1111/bdi.12089 |
| **Source type** | Review Article |
| **Keywords** | brain-derived neurotrophic factor; cAMP response element-binding protein; cytokines; hypothalamic-pituitary-adrenal axis; norepinephrine; protein kinase A; protein kinase C; serotonin; serotonin receptors; suicide. |
| **#Tags** | Biology, Suicidal Brain |
| **Summary of key points + notes (include methodology)** | Globally around 1 million induvials die of suicide every year. This means in the Unites States alone we lose 30,000 induvial. Possible risk factors include mental illness like mood disorders. Roughly 60% of suicides are linked to psychiatric illnesses occurring in induvial with mood disorder. Genetics are also increasingly prevalent with mood disorders and suicidal behavior increased risk. Not everyone with depression or psychiatric conditions are suicidal, however induvial that have intent to harm themselves or complete the act are suicidal. Initially they followed suicidal patients and compared their values to those who later died. Through this they focused on serotonin and norepinephrine systems, signaling pathways, neurotrophins (like BDNF), stress regulating HPA axis and immune function which show the most consistent findings. The direct access to human brain is very limited so the paper focused on indirect sources such as cerebrospinal fluid, blood, or postmortem tissue. The blood cells specifically lymphocytes (white blood cells) which interact with the immune system/brain and platelets which are widely used key features in neuron (Serotonin transporter protein is identical in both platelets and brain cells which |

| | |
|---|---|
| | means platelets have been used to measure serotonin receptors, monoxide activity and BDNF levels). Serotonin abnormalities in the 5HT system have been linked to suicidal thoughts and actions through measurements of serotonin/main metabolite 5-hydroxyindoleacetic acid (5HIAA) in the CSF and blood; postmortem brain studies; analysis of serotonin receptor subtype in blood platelets of suicidal patients; and neuroendocrine challenge tests that probe serotonin function. Through these measurements we were able to find that suicide behavior often has lower CSF 5HIAA and increased 5HT2A receptors (the metabolites). This was founded from the study in Asberg in 1976 where they discovered that patients with depression who attempted suicide had significantly lower CSF 5HIAA levels. Furthermore, blood studies showed lower serotonin in plasma and platelets than the healthy controls. There is higher 5HT1A receptors in nonviolent suicide victims, whereas the 5HT2C receptor editing patterns pre-mRNA were abnormal (more than one edit in one site). There is also a lot of research on norepinephrine and how MHPG ( a metabolite) is inconsistent with some studies showing an increase while other studies show a decrease in the metabolite. Furthermore Tyrosine hydroxylase (TH) also has mixed results with receptors and proteins which leads to inaccurate correlations between suicide and norepinephrine. There are also BDNF and TrkB Deceits in suicide victims especially In the hippocampus which leads to weakness in the brain's ability to adapt, recover, and resist stress. Additionally, there is an increase. In CRF for HPA Axis Dysregulation and a decrease in CRF-R1 and glucocorticoid receptors which leads to chronic stress hyperactivation. There was also Immune system involvement in an increase in pro-inflammatory cytokines and suggests that suicide is in part due to inflammation in the brain. In conclusion suicide biologically centers around serotonin dysfunction, stress system overdrive, reduction in brain growth factors, and inflammation, with risk increasing when trait such as impulsivity vulnerabilities meet state stressors like HPA and immune dysregulation. |
| **Research Question/Problem/ Need** | Through this they focused on serotonin and norepinephrine systems, signaling pathways, neurotrophins (like BDNF), stress regulating HPA axis and immune function which show the most consistent findings. |
| **Important Figures** | Ghanshyam N Pandey (Author) |

**Table 2**

Summary of studies of serotonin-2A receptors in postmortem brain tissue in suicide and depression

| Radioligand | Type of binding study | Brain region | Result | Reference |
|---|---|---|---|---|
| [$^3$H]spiperone | Homogenate | PFC 8, 9 | ↑ $B_{max}$ | Stanley and Mann 1983 (164) |
| [$^3$H]ketanserin | Homogenate | PFC 8, 9 | No change | Owen et al. 1983 (165) |
| [$^3$H]ketanserin | Homogenate | PFC 10 | No change | Crow et al. 1984 (166) |
| [$^3$H]ketanserin | Homogenate | Frontal cortex | No change | Owen et al. 1986 (167) |
| [$^3$H]spiperone | Homogenate | PFC 8, 9 | ↑ $B_{max}$ | Mann et al. 1986 (82) |
| [$^3$H]ketanserin | Homogenate | PFC 10 | No change | McKeith et al. 1987 (168) |
| [$^3$H]ketanserin | Homogenate | PFC 10, hippocampus | No change in PFC, ↓ in hippocampus | Cheetaham et al. 1988 (169) |
| [$^3$H]spiperone | Homogenate | PFC 8, 9 | ↑ $B_{max}$ only in violent suicides | Arora and Meltzer 1989 (170) |
| [$^{125}$I]LSD | Homogenate and sections | PFC 9 | ↑ $B_{max}$ and ↑ in sections | Arango et al. 1990 (83) |
| [$^3$H]ketanserin | Homogenate and sections | PFC, hippocampus | ↓ $B_{max}$ and ↓ in sections in PFC only | Gross-Isseroff et al. 1990 (85) |
| [$^3$H]ketanserin | Sections | PFC 9 | ↑ mid-layers | Yates et al. 1990 (171) |
| [$^3$H]ketanserin | Homogenate | PFC 10 | ↑ $B_{max}$ | Laruelle et al. 1993 (172) |
| [$^3$H]ketanserin | Homogenate | PFC 9, amygdala | ↑ $B_{max}$ | Hrdina et al. 1993 (173) |
| [$^{125}$I]LSD | Sections | Temporal and entorhinal cortex, hippocampus | No change | Joyce et al. 1993 (68) |
| [$^3$H]ketanserin | Homogenate | PFC 9, 10, 11 | No change in violent or nonviolent | Arranz et al. 1994 (174) |
| [$^3$H]spiperone | Homogenate | PFC 10, hippocampus | No change | Lowther et al. 1994 (175) |
| [$^3$H]ketanserin | Sections | PFC 10, hippocampus | No change | Stockmeier et al. 1997 (176) |
| [$^3$H]ketanserin | Homogenate | PFC 8, 9 | ↑ $B_{max}$ | Turecki et al. 1999 (177) |
| [$^3$H]ketanserin | Homogenate | PFC 9, 10, 11, hippocampus | No change in PFC, ↓ $B_{max}$ in hippocampus | Rosel et al. 2000 (178) |
| [$^3$H]LSD | Homogenate | PFC 8, 9 | ↑ $B_{max}$ | Pandey et al. 2002 (23) |

PFC = prefrontal cortex; ↑ = increase; ↓ = decrease.

Table 2 is on the summary of studies of Serotonin-2A recptors in the postmortem brain tissue in suicide and depression.

**VOCAB: (w/definition)**

Lymphocytes: Essential for immune system white blood cells, important for fight9ng infection and disease by recognition, remembering foreign pathogens and targeting in viruses and bacteria.
Neurotrophins: Family of growth factors crucial for development, survival, and function of nerve cells (neurons) in both the central and peripheral nervous systems.
Norepinephrine: Also known as noradrenaline is a naturally occurring chemical that functions as both a hormone and a neurotransmitter

**Cited references to follow up on**

Beck AT, Steer RA. Clinical predictors of eventual suicide: a 5- to 10-year prospective study of suicide attempters. J Affect Disord. 1989;17:203–209. doi: 10.1016/0165-0327(89)90001-3. [DOI] [PubMed] [Google Scholar]

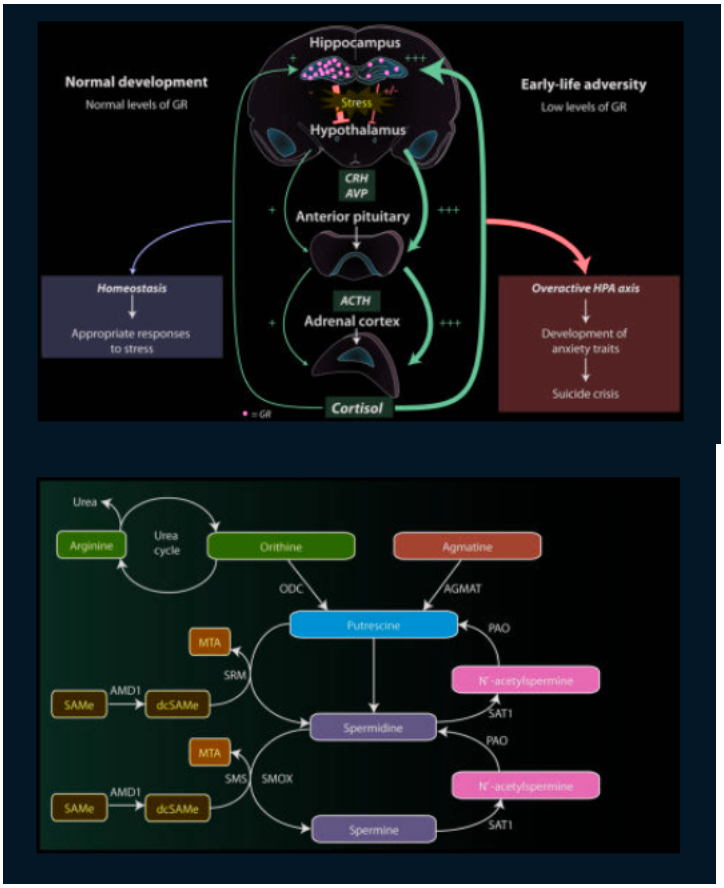
**Follow up Questions**

How can suicide be predicted?

# Article #10 Notes: The molecular Basis of the Suicidal Brain

| | |
|---|---|
| **Source Title** | The molecular bases of the suicidal brain |
| **Source citation (APA Format)** | Turecki, G. (2014). The molecular bases of the suicidal brain. *Nature Reviews Neuroscience*, *15*(12), 802–816. https://doi.org/10.1038/nrn3839 |
| **Original URL** | https://www.nature.com/articles/nrn3839 |
| **Source type** | Review Article |
| **Keywords** | Suicidality, Distal Risk Factors, Early Life Adversity, Proximal Risk Factors |
| **#Tags** | Molecular Science, Suicide, Biology |
| **Summary of key points + notes (include methodology)** | Suicide is one the leading causes of death worldwid, especically in young children due to the lack of effective treatemtns. Due to the stigma and lack of awareness around suicide indicuals who are at risk often do not seek help, making suicide a “silent epudemic”. The phrase suicidlity covers a whole spectrum of suicide including suicidal thoughts, attempts, and completed suicide (deaths). Most indivuals who commit or die due to suicide are effected with psychiatric disorders particularly depression, schizophernia, substance abuse, or personility disorder. The researchers modeled suicide risk as the interaction of distal, long term predisposing, and prozimal, short term triggering, factors. The distal factors increase vulnerbility across life while proximal factors predict a crisis. Distal (Predisposing) Factos are found in family genetics, showing suicide and how it runs in families, not simply because psychiatric disorders are heritiable but also due to the fact that suicidal behavior itslef can be transmitted through generations. Moreover this is seen in genetic variation with candidate gene studies showing potential variants of serotonin transporter, tryptophan hydroxylase, and MAOA and similarities between them (although they have been inconsistent results). Early-life Adversity (ELA) is also a distal factor that plays a huge role in suicide, with instances such as child abuse and trauma, with the closer the abuser and the more frequent the abuse the stronger the risk. Further more the biology behind ELA would be epigentic changes (in the DNA and methylation) which modify gene expression long-term. Moreover the hypothalamus-pituaruary adrenal (HPA) axis is overactive in indivuals exposed to ELA leading to heightened |

| | |
|---|---|
| | stress sensitivity. Studies have also showed methylartion changes in the gluccocorticoid recptor gene (. NR3C!), which impaires stress regulation. Other epigentic changes include affects on stress hormone responses (FKBP5), and neuronal growth and plasticity (BDNF and TRKB). Also, animal models have confirmed that poor maternal care or early stress can elad to lasting DNA methylation shifts and abnormal stress repsonses, which leads to epigenetic modification that are stable over time explaining how childhood trauma can influence suicide risk decades later. Furthermore there are mediating factors such as distal risk which act through traits like impulsivity, aggression, and anxiety (partly heritable and shaped through early environment). Also, families with higher impulsivity/aggressive traits show greater clustering of suicide which strongly correlate with suicidality in both population and clinical studies. Substance abuse is also a factor that can lead to suicide risk. Low serotonin and low cholesterol levels are linked with impolite and aggression as well. Finally proximal (triggering) factors include life stressors with a combination of psychiatric and environmental episodes. Almost all suicidal deaths show psychiatric symptoms the months leading up to the death. Moreover, a depressive state (marked by hopeless and impaired judgment) is strongly linked to suicidal crisis. Molecular changes in the brian during a crisis include serotonin deficits (low 5-HIAA in CSF, reduced transporter/recptor avalability), polyamine system alteration (regulates stress responses), glutamate and GABA dysregulation (effectiveness of rapid-acting glutamatergic antidepressants), inflammatory changes (higher IL-6 and lower IL-2 levels in suicidal indivuals), Glial dysfunction (reduced astrocytic support), and HPA axis abnormalities (increase in CRH, altered glucocorticoid receptor function and enlarged adrenal glands). Overall suicide is heterogeneous with more precise modesl needed to separate suicide-specific biology from general psychiatric pathology. Better molecular understanding could guide improved prevention and personalized treatment strategies. |
| **Research Question/Problem/ Need** | The review focuses on how biological and molecular processes link the risk factors impact suicidal behavior. |
| **Important Figures** | Gustavo Turecki (Author) |

| | |
|---|---|
| |  <br>Figure 1 shows the early-life adversity cuases low glucocorticoid recptor levels leading to an overactive HPA axis and increased stress and suicidal risk. Figure 2 shows the polyamine biosynthesis pathways where arginine and ornithine are converted into other moelcules which are essential for cell growth and function. |
| **VOCAB: (w/definition)** | Endophenotypes: Traits that associate with an illness in the population, are heritable, state independent, and co-segregate with the condition investigated.<br>Glucocorticoid: A class of steroid hormones that play a crucial role in body's physiological and immune responses.<br>State Markers: Biological, Psychological, behavioral, or clinical markers associated with a given phenotype. |
| **Cited references to follow up on** | Brent, D. A., Bridge, J., Johnson, B. A. & Connolly, J. Suicidal behavior runs in families. A controlled family study of adolescent suicide victims. *Arch. Gen. Psychiatry* **53**, 1145–1152 (1996)<br><br>Perlis, R. H., Ruderfer, D., Hamilton, S. P. & Ernst, C. Copy number variation in subjects with major depressive disorder |

| | |
|---|---|
| | who attempted suicide. *PLoS ONE* **7**, e46315 (2012). |
| **Follow up Questions** | The relationship with passed down gentic traits for psychiateric disorders and how it relates to suicide. Are these psychiatric genetics that are passed down related to psychiatric disorders being a risk factor. |

# Article #11 Notes: CRISISS: A Novel, Transcriptionally and Post-Translationally Inducible CRISPR/Cas9-Based Cellular Suicide Switch

| | |
|---|---|
| **Source Title** | CRISISS: A Novel, Transcriptionally and Post-Translationally Inducible CRISPR/Cas9-Based Cellular Suicide Switch |
| **Source citation (APA Format)** | Amberger, M., Grueso, E., & Zoltán Ivics. (2023). *CRISISS: A Novel, Transcriptionally and Post-Translationally Inducible CRISPR/Cas9-Based Cellular Suicide Switch*. International Journal of Molecular Sciences, 24(12), 9799–9799. https://doi.org/10.3390/ijms24129799 |
| **Original URL** | https://www.mdpi.com/1422-0067/24/12/9799 |
| **Source type** | Research Article |
| **Keywords** | CRISPRR/Cas9, suicide switch, inducible system, gene therapy safety, transcriptional regulation, post-translational control, synthetic biology |
| **#Tags** | CRISPR, GeneTherapy, SyntheticBiology, CellularSafety, InducibleSwitch |
| **Summary of key points + notes (include methodology)** | As gene and cell therapy technology begins to grow and become more prevalent in medicine, there has been a strong need for safety to control life-threatening side effects. The study introduced a system called CRISPR-Induced Suicide Switch (CRISISS) – which is a method that can destroy genetically modified cells in a controlled and efficient way. CRISISS works by using a Cas9 enzyme to cut Alu DNA segments (Highly Repetitive DNA fragments in the human genome) – cutting these segments causes massive irreversible DNA damage leading to cell death. The system includes a Cas9 enzyme and an Alu-specified guide RNA that directed the Cas9 where to cut. The components were induced into the cell using a Sleeping Beauty Transposon system (which allows stable integration into the genome). The genetically modified calls showed no harmful effects when CRISISS was inactive, with no background Cas9 activity, no DNA damage, or unexpected cell death. However, when Cas9 was turned on there was lots of DNA damage immediately and the cells died within four days. |

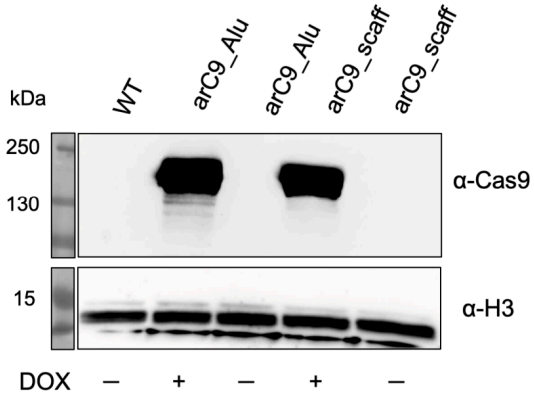
Modern genetic advances have shown new openings in adding new genes or altering existing ones, with theses engineered cells acting as “living drugs”, capable of treating diseases that were previously incurable. An example of this would be the adoptive T-cell therapy which is particulally called CAR-T cell therapy which has received approval from the FDA and EMA. Several products are already being used to treat patients however these treatments involve inserting new genetic material into a cell’s genome which can often accidently trigger disruption of important genes, change how genes are turned on and off, and cause harmful side effects including cancerous transformations. One clinical study with CAR-T cells showed random integration that ked to lymphoma formation in some patients. In addition serious side effects such as neurotoxicity and cytokine release syndrome (CRS) can make treatment dangerous. With other similar concerns with dingle-gene disorders or regenerating tissue using induced pluripotent stem cells, scientists decided to find a way to have built in safety systems. A suicide switch is a genetic safety feature that allows scientists to permentently remove modified cells from patients. Usually the suicide switch in inserted into the same vector as the therapeutic so every modified cell has both the therapeutic and the suicide switch. Current switches work differently:

- Some activitate apoptosis genes such as indusable caspase-9
- Others use enzyme -podrug systems such as Herpes Simplex vius thymidine kinase (HSV-TK), or cytosine deaminase (CD) which converts harmless drugs into toxic molecules
- Another system uses cell-surface markers allowing immune antibodies to recognize and kill the modified cells

The old systems have multiple drawbacks including the failing to kill modified cells, prodrugs can have toxic side effects, systems can be recognized by immune systems and attacked, some rely on the cell cycle, and they can worsen the cytokine release syndrome.

The new system, CRISISS that is a stable, inducible and highly effective way at killing modified cells, used as a tool to destroy the genome of target gets in Alu retrotransposons (repetitive DNA sequences that make up a large part of the human genome . The system was conctrucgted as a multi-compnont system using the Steeping Beuty Transposon for stable integreation, it has four main components. 1. Alu-specif single-guide RNA sgRNA – controlled by human U6 promoter RNA guude to Alu),, 2. Allosetrically switchable Cas9 (allows activation by 4-HT), 3. Reverse Transactivator (Tet-On M2, expressed in PGK promoter, protein binds to the TRE and activitates Cas9 expression in the presence of DOX), 4. Neomycin Resistence gene (allows for selction of cells that have successfully integrated the CRISISS construct. It works by adding doxycycline which turns on the Cas9 gene expression (transcriptional

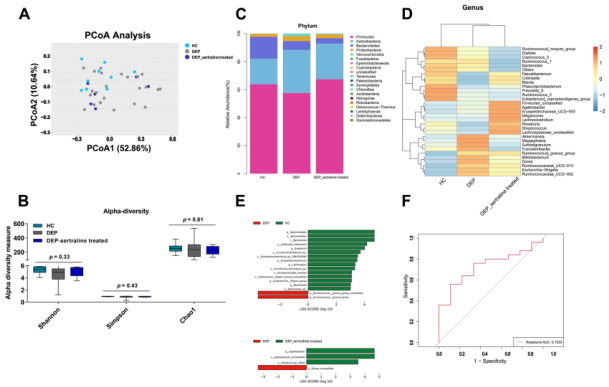
| | |
|---|---|
| | control) then by adding 4-HT which activated the Cas9protien (post translational control). The active cas9 is buided by a sgRNA and makes thousands of double strand DNA breaks within the genome, with maximizing the DNA damage and ignoring the normal off-targer concerns.To test the function of the system researchers inserted the HeLa cell lines that contained the full CRISISS construct , they used Sleeping Beauty transposon system to intert entire CRISISS cassette into the cell's DNA in a stable way, two main versions: 1. CRISISS-Alu cells – contained both the Cas9 components and Alu-specific guide RNA, 2. CRISISS-sgRNA cells – contain Cas9 components but no guide RNA (control). Both had the SB100X transposase enxyme to help with stable integrationa dn cells that se\uccessfully recived the contruct were sekected using antibiotic G418. Initially they wanted to make sure Cas9 was only produced when DOX was added so they exposed CRISISS cells to different concentrations of DOX, collected cell sampels after 48 hours and checked for Cas9 protien which resulted in the fact that without DOX, Cas9 was undetactable showing there was no background "leakiness", DOX was added Cas9 expression increased in a dose dependent manner, and expression levels stayed high and stable for several days after induction. The transcriptional control fo Cas9 through TRE promoter and Tet-On system were working, however Cas9 remained fully inactive until DOX was present. Next the reseafchers tested whether the Crisiss actually caused DNA damage by monitoring the DNA damage using a marker called phosphorlylated KAP1 which appears when cells experience double-strand DNA breaks. The setup was that the cells were divided into four groups, 1. No induction, 2. DOX only, 3. 4-HT only, 4. Both DOX and 4-HT, with both the CRISISS-Alu and CRISISS-sgRNA cells tested. Only the CRISISS-Alu cells in group four showed strong pKAP1 signals indicating major DNA damage. Next step was to show how this affected cell survival and growth. They used the same ecperiemntal groups and the cells were monitered for cell morphology (appearance), proliferation rate, and viability. Observed that CRISISS systems were succesfuly caused massive cell death and complete growth arrest but only in the preseance of sgRNA and DOX, and the presence of guide RNA. Over all the CRISISS system wokrs only when riggered by both inducers, casuses massive DNA Damage within 4 days but has no background or leaking damage. Methodology: Used plasmid construction with a SB vector backbone in various cultures of HeLa human celss to integrate the sucide switch with different factors. Used cell growth and viability tests such as the growth curve assay to test the amount of growth and colony formation assay with variation in the test groups mentioned above. Used student's t-tst with a p value threshold of $p<0.05$. |
| **Research Question/Problem/** | The study talks about the need for a controllable and safe "suicide switch" |

| | |
|---|---|
| **Need** | in engineered cells, such as CAR-T cells, to prevent uncontrolled effects of off-target activity. |
| **Important Figures** |  <br>Fig 3. Dose-response curves for small-molecule inducers of suicide switch |
| **VOCAB: (w/definition)** | CRISPR/Cas9: Genome-editing tool that introduces targeted DNA cuts guided by an RNA sequence<br>Suicide Switch: A genetic system engineered to induce cell death under controlled conditions<br>Transcriptional control: regulation of gene expression at the RNA synthesis level<br>CAR-T cells: Chimeric antigen receptor T cells engineered to target cancer cells |
| **Cited references to follow up on** | Micklethwaite, K.P.; Gowrishankar, K.; Gloss, B.S.; Li, Z.; Street, J.A.; Moezzi, L.; Mach, M.A.; Sutrave, G.; Clancy, L.E.; Bishop, D.C.; et al. *Investigation of product-derived lymphoma following infusion of piggyBac-modified CD19 chimeric antigen receptor T cells*. Blood 2021, 138, 1391–1405<br><br>Wang, X.; Chang, W.C.; Wong, C.W.; Colcher, D.; Sherman, M.; Ostberg, J.R.; Forman, S.J.; Riddell, S.R.; Jensen, *M.C. A transgene-encoded cell surface polypeptide for selection, in vivo tracking, and ablation of engineered cells*. Blood 2011, 118, 1255–1263.<br><br>Oakes, B.L.; Nadler, D.C.; Flamholz, A.; Fellmann, C.; Staahl, B.T.; Doudna, J.A.; Savage, D.F. *Profiling of engineering hotspots identifies an allosteric CRISPR-Cas9 switch*. Nat. Biotechnol. 2016, 34, 646–651 |
| **Follow up Questions** | How does CRISISS compare to the inducible suicide switch systems like iCasp9 in the way the response to speed and leakiness?<br>How would the off-target effects of CRISPR/Cas9 influence the safety of CRISISS system? |

# Article #12 Notes: Microbiome and tryptophan metabolomics analysis in adolescent depression: roles of the gut microbiota in the regulation of tryptophan-derived neurotransmitters and behaviors in human and mice

| | |
|---|---|
| **Source Title** | Microbiome and tryptophan metabolomics analysis in adolescent depression: roles of the gut microbiota in the regulation of tryptophan-derived neurotransmitters and behaviors in human and mice |
| **Source citation (APA Format)** | Zhou, M., Fan, Y., Xu, L., Yu, Z., Wang, S., Xu, H., Zhang, J., Zhang, L., Liu, W., Wu, L., Yu, J., Yao, H., Wang, J., & Gao, R. (2023). *Microbiome and tryptophan metabolomics analysis in adolescent depression: roles of the gut microbiota in the regulation of tryptophan-derived neurotransmitters and behaviors in human and mice.* Microbiome, 11(1). https://doi.org/10.1186/s40168-023-01589-9 |
| **Original URL** | https://link.springer.com/article/10.1186/s40168-023-01589-9 |
| **Source type** | Research Article |
| **Keywords** | 5-HT (Serotonin), Kynurenine (Kyn), TPH1/TPH2, IDO1/3HAO, FMT, CRS Roseburia Intestinalis |
| **#Tags** | AdolescentDepression, GutMicrobiota, TryptophanMetabolism, Neurotransmitters |
| **Summary of key points + notes (include methodology)** | Adolesant depression is a prevelatn issue in this up and coming world, often starting from a teenager and leading into adlt causing serous emotional and functional damge. Traditional antidepressant medication is not always effective so scientists are looking for biological factors that contribute to depression. The gut-brain azix (GBA) with connections between the guy microbiome and brain function has become increasingly prevelant. The study focuses on how tryptophan (Trp) an amino cid that produces seotonine (5-HT) and kynurenine (Kyn) are effected by gut bacteria with imbalnaces in the T-s-k pathways influencing mood and behavior. The study aimes to compare the gut mircobiodata between helathy and depressed adolesants, test whether transferring healthy gut bacteria to mice can reduce deprepression like behavior, and study how Roseburia intestinalis affects neurotransmitters like serotonin and kynurenine in mice. The study deisgn overview includes a human clinical study to find how gut microbiota and tryptophan metabolism differ between depressed and healthy adolescents and to test if these microbiota diffrences can cause depression-like bevaiors in mice and identify which |

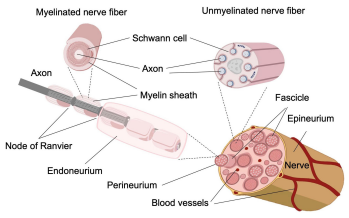
| | |
|---|---|
| | bacteria are responsible. They combined microbiome sequencing, metabolomics (measures of tryptophan and its metabolites), fecal microbiota tranplantaiton, and molecular biology assays (gene expression, enzyme activity and histology. The human study deisgn had 35 adolesants, with 25 diagnosied with MDD, and 10 healthy with inclusion criterias being no antibitics or probiotics in the last 3 moths and no chronic diseases. Exclusions incuded neulogical, endorcirne, or gasteroninstenal diseases. The depression severity was measured with the Revised Child Naxiety and Depession Scale and had antidepressant therapy with sertranline (SSRI) for 8 weeks with follow up sampeles of fecal,urinal, and blood samples for mircrobiome sequencing, plasma metablomics, and metabolic validation. They went through mircrobiome analysis then used the metabolomics (such as tryptophan, serotonin, kynuerine, and more) to find staticial correlations with bacterial abundances and depressions scores. They used mouse models of depression of chronic resistraint stress (CRS) for 28 days to induce depression. The fecal microbiota (FMT) was used to determine whether gut bacteria from dpressed adoloscents can cause deoressie behavior in mice with four groups, who were transplaed with a broad spectrum antibiotics for 5 days to clear native microbiota. The used the Sucrose preference test, tail suspensiton test, forced swin test, and open field test to measure depression-like behavior in mice. After this the mice were used to test the neurotransmitters (ie brain and color analyzed for serotonin and the other moelcules), enzyme expression, inflammation and neuronal health and gut barrier intergrity. The roseburia bacteria was essential to test whether the specific bacterium toseburia can repsorduce theantidepressant effect of the healthy microbiota through CRS mice that were orally give Roseburia while the control mice received vehicle solution. They ised the same behavioral tests then also tested the metabolites and enzyme levels as well as gut tissue finding that the Rosbureia reviesed depression like begavior, increased serotonin, reduced toxic metaboloties, and restored gut and brain structure integrity. Used software SPSS v25, grapjhpaf Prism 9, and R and tests used were one/two way ANOCA followed by Tukey post hoc and other stattictical and computer anlysis. In the human study they found that depressed teenagers had a significanly different gut microbial composition compared to healthy controls. Alpha diversity (the number of species and eveness) was loves in MDD patients which indivuared reduced microbiome richness. The beta diversity (PCoA) showered clear seperatin between two groups. Further more specific bateria were reduced in depression candiates, for example indivuals with higher depression had lowed Roseburia, however with sertraline treatment over 8wekk the Roseburia increased. There were also changes in tryptophan metabolites with the t-s-k pathway strongly affected. The mouse experiment with the CRS mise showered reduced sucrose |

| | |
|---|---|
| | preference and increased immbolity and reduced activity. In the FMT (fecal microbiota transplant) the mice that recived depressed microbiota developed/maintained depression like symptoms similar to CRS mice. The mice that received healthy microbiota showed major behavioral improvement, with conclusion ebing that gut microbiota from depressed adoleants can transfer depressive behavior to mice while halthy microbiota can rescue it. The roseburia intential supplementation leads to colonization and teolerability, the behavioural outcomes were also better, showing that the loss of roseburia in adolesants disrupts trptophan metabolism, reduces serotonin, increases toxic metabolies and contributes to depression. When Rosburia is reintroduced the serotonin levels strengthen and the gut-brian bairreri and reverse the depressive behaviors. |
| **Research Question/Problem/ Need** | The study aimes to compare the gut mircobiodata between helathy and depressed adolesants, test whether transferring healthy gut bacteria to mice can reduce deprepression like behavior, and study how Roseburia intestinalis affects neurotransmitters like serotonin and kynurenine in mice. |
| **Important Figures** |  <br>Fig 1. Differences in gut microbiota composition between depressed and healthy adolescents |
| **VOCAB: (w/definition)** | Tryptophan: An essential amino acid that serves as a precursor for serotonin and other neurotransmitters<br>Metabolomics: The large-scale of metabolite within cells, tissues, or organisms<br>Fecal Microbiota Transplantation (FMT) Transfer gut microbiota from a donor to recipient to study/treat microbiome-related conditions<br>Adolescent Depression: A mental health disorder in teenagers characterized by persistent sadness, irritability, and behavioral changes |
| **Cited references to follow up on** | Kelly, J. R., et al. (2016). *Breaking down the microbiome–gut–brain axis in depression*. Nat Rev Neurosci, 17, 60–73<br>Valles-Colomer, M., et al. (2019). *The neuroactive potential of the human* |

| | |
|---|---|
| | *gut microbiota in quality of life and depression*. Nat Microbiol, 4, 623–632. |
| **Follow up Questions** | Which specific gut microbial taxa are most strongly associated with altered tryptophan metabolism in depressed adolescents?<br>What are the limitations of translating the findings in a mouse to a human adolescent? |

# Article #13 Notes: Use of an invertebrate animal model (*Aplysia californica*) to develop novel neural interfaces for neuromodulation

| | |
|---|---|
| **Source Title** | Use of an invertebrate animal model (Aplysia californica) to develop novel neural interfaces for neuromodulation |
| **Source citation (APA Format)** | Zhuo, J., Gill, J. P., Jansen, E. D., Jenkins, M. W., & Chiel, H. J. (2022). *Use of an invertebrate animal model (Aplysia californica) to develop novel neural interfaces for neuromodulation*. Frontiers in Neuroscience, 16. https://doi.org/10.3389/fnins.2022.1080027 |
| **Original URL** | https://www.frontiersin.org/journals/neuroscience/articles/10.3389/fnins.2022.1080027/full |
| **Source type** | Review Article |
| **Keywords** | Aplysia Californica, neural interface, neuromodulation, invertebrate model, electrophysiology, nervous system |
| **#Tags** | Nervous System, Neuromodulation, Aplysia Californica, Invertebrate model, electrophysiology |
| **Summary of key points + notes (include methodology)** | The articles summarizes how the animal aplysia californica serves as a valuable model organism for developing testing neural interface technology and use in neuromodulation through methods to record, stimulate, or inhibit activity. Aplysia is very useful because of its neurons that are large and identificaly and electrically compact and its nerves are unmyelinated making them idela for studying the unmyelined C-fibes in vertabrates (organism that transmit pain and sesonry signals). There was also a strong emphasize on the new neurpmodeulation tools like high frequency alternating current and more and how it depends on the understanding f funcdamental biophysical processes. The organism has unmyelinated azons which are about 10 micrometers in diamet and mirror vertebreate C fibers that carry pain and sensory signals. Moreover they have longevity in Ex vivo conditions which means that their neural tissues remain viable for long hours outside theyr body because the animal antually tolerate envimental changes in temperature. Further more they have axon length and identification with long peripheral nerves whch allow sepration of axons by conduction velocity. Finally they also ahev large and identificable neurons that allow for easier electirbility conditons and testing. Promotion techniques also have developed and tested and afflict CIA with an increase in electrical and hybrid stimulations making them easier to work with and |

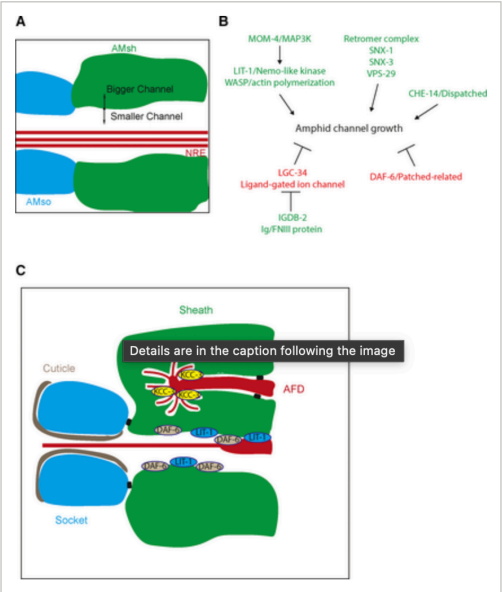
| | |
|---|---|
| | mechanical modulations such as focused ultrasounds the Organism allows researchers to systematically study excitation inhibition without the complexity of vertebrae brain furthermore we can use optical techniques such as infrared neural inhibitions which allow us to test nerves and nerve pulses within the brain can also use mechanistic studies which show thermal acceleration of voltage gated potential potassium channels which could lead to action potential education and how this could lead to suicide in my project finally the overall synthesis of the findings show us that with these organisms we can use infrared neural inhibition which is proven to selectively and reversibly block small diameter on myelinated axons both in single identified neurons at the compound nerve level we can use it as a mechanistic insight to see the action potential and the temperature induced acceleration of potassium channel kinetics without sodium channel blockade we can see selective demonstration of large diameter axons versus the continued firing of small diameter axons which could be blocked and we can see the translation potential which is the I and I protocol which developed in this Organism were successfully applied to mammals with vertebrae so some key advantages to using this error Organism is the biological simplicity comparative cytological relevance to vertebrates and their unmyelinated fibers furthermore they are reusable long lasting neural tissue which enables us to test them for a long time there no neuroin identity and accessibility for precise control and their range of Axon diameter are ideal for testing size selective neuromodulation in conclusion this review looks at the most powerful aspects of why this Organism can be used for developing and validating neural interface technology with aims of neuromodulation. |
| **Research Question/Problem/ Need** | The study investigates how the model organism can be used to develop and test new neural interfaces in neuromodulation, to make a simpler system to study neural circuitry. |
| **Important Figures** |  <br>FIGURE 1<br>Schematic of the structure of a peripheral nerve (Guedan-Duran et al., 2020. The original work is published with permission under a Creative Commons Attribution 4.0 International License. To view a copy of this license, visit http://creativecommons.org/licenses/by/4.0/).<br>Fig 1. shows the anatomy of the aplysia californixa nervous system highlighting ganglia used for neural interface experiments. |
| **VOCAB: (w/definition)** | Neuromodulation: Therapeutic alteration of nerve activity through targeted |

| | |
|---|---|
| | electrical or chemical stimulation<br>Electrophysiology: Study of electrical properties of biological cells and tissues<br>Ganglion: a collection of nerve cell bodies in the peripheral or central nervous system |
| **Cited references to follow up on** | Lertmanorat, Z., and Durand, D. M. (2004). *A novel electrode array for diameter-dependent control of axonal excitability: A Simulation study*. IEEE Trans. Biomed. Eng. 51, 1242–1250. doi: 10.1109/TBME.2004.827347<br><br>Regehr, W. G., Pine, J., Cohan, C. S., Mischke, M. D., and Tank, D. W. (1989).*Sealing cultured invertebrate neurons to embedded dish electrodes facilitates long-term stimulation and recording*. J. Neurosci. Methods 30, 91–106. doi: 10.1016/0165-0270(89)90055-1<br><br>Smarandache-Wellmann, C. R. (2016). *Arthropod neurons and nervous system.* Curr. Biol. 26, R960–R965. doi: 10.1016/j.cub.2016.07.063 |
| **Follow up Questions** | Are there specific types of neuromodulation therapies that would benefit most from insight gained using this model? |

# Article #14 Notes: *C. elegans* as a model to study glial development

| | |
|---|---|
| **Source Title** | C. elegans as a model to study glial development |
| **Source citation (APA Format)** | Zhang, A., & Yan, D. (2021). *C. elegansas a model to study glial development.* The FEBS Journal. https://doi.org/10.1111/febs.15758 |
| **Original URL** | https://pubmed.ncbi.nlm.nih.gov/33570807/ |
| **Source type** | Research article |
| **Keywords** | C. elegans, glial cells, neurodevelopment, model organism, nervous system, genetic regulation, glial differentiation |
| **#Tags** | C.Elegans, Glial development, neurobiology, nervous system |
| **Summary of key points + notes (include methodology)** | Glia make up about half of the cells in the mammalian nervous system. Initally they were throught to be passive support cells for neurons however now we know them to be functional and morphologixaly diversive and incolve synapse formation, pruning, ion balncace, neurotramitter recyclying, BBB maintaince, and neural circuit regulation. Despirte this information there there many unknown knowledge gaps with the three most important being glial diversity and how it is generated, morphogenesis and how it occurs, and finally how developmental disruptions lead to neurological diseases. Challenges in studying glia would be that mamalians brain contain many hetreogenious glial subtypes and it is hard to keep track of them, furthermore multiplative glia in vivo often elads to elathlity since they are essential for neuron survival, moreover in vitro glia loose their native interations with neurons and other glia which limit insights. C. Elegans allow a simplified, genetically tractable model which is transparent body for live imaging, the entire cell linage is mapped, glia are not essential for survival, allowing for function manipulation, and the nerveosu system contains 302 neurons and 50 ectodermal glia. C. Elegans are hemaphrodite worms with 302 neurons, 50 ectoderm-derived glia, and 6 mesoderm-deriveled glial like cells. The extrodermal cells are Two different types of cells including sheet cells and socket cells furthermore cincilla which are sensory organs allow glial form channels which are used to guide sensory dendrites into the environment for example amplified sheath and socket glia in the head allow and sheathing of sensory dendrites forming channels that allow neural environmental interactions furthermore cephalic sheath glia extended into the posterior of the nerve ring so glia across the elegant sense organs have distinct |

| | |
|---|---|
| | morphology and gene expression profiles even glia which performed several similar roles have very distinct and different structure and molecular identification which I think goes back to the biological idea of form fits function the key transcription factors regulate geophyte specification and diversification so they work as a signal transduction pathway I truly believe so other major genetic regulations have glial faith include gene proteins such as Lin 26 ALR one pros one and others that kind of just regulate these neural network furthermore there are subtype specific promoters which allow equalization and manipulation of individual glial populations and their single cell RNA sequencing which map transcriptional profiles for specific glia. Furthermore you can see under stress worms enter a dower stage and there's also examples of morphological plasticity which is response to environment there is squeal process extensions which is retrograde extensions their compartmentalization ion homeostasis and so broadly key findings include that sociology Anglia exhibit cell type specific genetic programs that are highly conserved with vertebrate glia genesis furthermore transcriptional regulation God's glial specification diversification and maintenance and then the morphogenesis of the glial structures and the animals depends on the vesicle transferring misguided skeleton remodeling the adhesion proteins and the neural signal furthermore the neuron glia interactions are reciprocal so the neuron influences glial shape while the glia shape influences the neural receptive ends and so this kind of goes back to that form fits function and then glia shows developmental and environmental plasticity which means that it can confuse or it can break apart due to environmental or external stressors and so overall C elegans provide to be a very powerful and genetic live image model for glial biology and they're very simple so it allows us to use their glia and we can tweak the glia without harming the animal itself and feeling overall glia served as an accessible and genetically tractable system to uncover universal principles about glia and glial cells. |
| **Research Question/Problem/ Need** | The study explores how C. elegans ca be used as a model organism to research the development, differentiation, and function of glial cells |

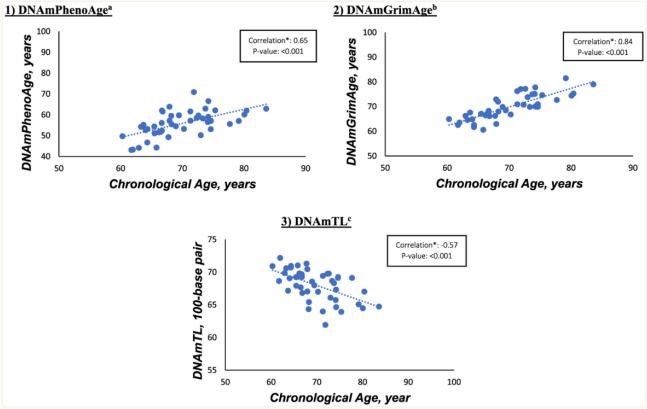
| | |
|---|---|
| **Important Figures** |  <br>Genetic Pathways regulating glial development and differentiation |
| **VOCAB: (w/definition)** | Glial cells: non-neuronal cells in the nervous system that provide support protection, and modulation of neurons<br>Differentiation: The process by which a less specialized cell becomes a more specialized cell type<br>Genetic Tractability: the ease with which an organism's genes can be manipulated for experimental purposes<br>Neurodevelopment: The process by which the nervous system grows and develops |
| **Cited references to follow up on** | Bayraktar OA, Fuentealba LC, Alvarez-Buylla A & Rowitch DH (2014) *Astrocyte development and heterogeneity*. Cold Spring Harb Perspect Biol 7, a020362.<br><br>Chung WS, Welsh CA, Barres BA & Stevens B (2015) *Do glia drive synaptic and cognitive impairment in disease?* Nat Neurosci 18, 1539–1545.<br><br>Zuchero JB & Barres BA (2015) *Glia in mammalian development and disease.* Development 142, 3805–3809.<br><br>tork T, Bernardos R & Freeman MR (2012) *Analysis of glial cell development and function in Drosophila*. Cold Spring Harb Protoc 2012, 1–17. |
| **Follow up Questions** | Which specific genes in *C. elegans* are critical for glial development and differentiation?<br>What limitations exst when translating findings from *C. elegans* glia to mammalian glial biology? |

# Article #15 Notes: Pilot Study of Second-Generation DNA Methylation Epigenetic Markers in Relation to Cognitive and Neuropsychiatric Symptoms in Older Adults

| | |
|---|---|
| **Source Title** | Pilot Study of Second-Generation DNA Methylation Epigenetic Markers in Relation to Cognitive and Neuropsychiatric Symptoms in Older Adults |
| **Source citation (APA Format)** | Vyas, C. M., Sadreyev, R. I., Gatchel, J. R., Kang, J. H., Reynolds, C. F., Mischoulon, D., Chang, G., Aditi Hazra, Manson, J. E., Blacker, D., Immaculata De Vivo, & Okereke, O. I. (2023). *Pilot Study of Second-Generation DNA Methylation Epigenetic Markers in Relation to Cognitive and Neuropsychiatric Symptoms in Older Adults*. Journal of Alzheimer's Disease, 93(4), 1563–1575. https://doi.org/10.3233/jad-230093 |
| **Original URL** | https://journals.sagepub.com/doi/10.3233/JAD-230093?url_ver=Z39.88-2003&rfr_id=ori:rid:crossref.org&rfr_dat=cr_pub%20%200pubmed |
| **Source type** | Research article |
| **Keywords** | Alzheimer's disease; cognition; DNA methylation; epigenetics; neuropsychiatric symptoms |
| **#Tags** | Epigenetics, DNAMethylation, Cognition, Neuropsychiatry |
| **Summary of key points + notes (include methodology)** | The human brain contains hundreds of trillions of synapses which form changes and disappear dynamically across developmental and experiences so this entails individuals that grow tend to lose and gain multiple different synapses as they age. Synapse elimination also known as pruning is crucial for refining and perfecting neural circuits which ensure efficiency and information processing and maintenance of the brain homeostasis while neural mechanisms of synaptic activity information have been well characterized researchers now realizing that glial cells play an active and essential role in synapsis illumination and maintenance initially glial cells were not a support cells but now are recognized as active participants in the sculpting of the neural circuits by taking over and degrading synapses this engulfment process is also known as phagocytosis which I know it as cell Eden where glio recognizes internalizes and digests synaptic materials almost like awesome so proper glial engulfment ensures developmental refinement while defects can lead to neural development and neurodegenerative disorders such as Alzheimer's disease Parkinson's disease autism schizophrenia and many more major types of glia involved |

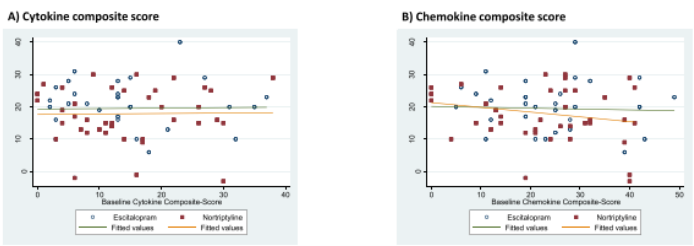
include microglia gum astrocytes and peripheral albium microglia or are seen in the CNS macrophages and they remove excess synapses during development the astrocytes however engulf and degrade synapses in both development and adulthood with different receptor systems such as MGF 10 and MERTK finally peripheral agria are Shawn cells in the PNS and glia and the model organisms like C elegans or drosophilia also engulf axons and synaptic remnants this review emphasizes how sympathetic engulfment biglia is conserved regulated in highly specific process across species it also helps us understand glial engulfment and how it offers a new insight into brain circuit formation and refinement the maintenance of synaptic health and the mechanism underlying brain disease and repair one experiment that allowed us to see this glial engulfment was a mouse and a mammal system which was used to study microglia in astrocyte mediated synaptic engulfment it was done through immunocytochemistry for synaptic and glial markers and electron microscopy for engulfed vesicles furthermore there have been studies on drosophilia and how their simple nerve system has been able to use identifiable glial subtypes such as cortexin glia and in cheap Anglia which engulf Axon terminals during development there are also been zebrafish studies which have shown live imaging of microglial engulfment of synapsis during the neural circuit refinement. So a very important molecular mechanism which underlies this engulfment is the recognition of the signals that the synapses release in order for the glial to detect and eventually engulf them this would be the PS or the phosphatidylserine and its exposure on the outer membrane which acts as a key signal in the signal transduction pathway normally the PS is on the inner leaflet and is flipped outward in distress or remodeling synapses glial receptors that recognize PS are the PSR one the Tam family receptors drapers and the CD one also bridging molecules which are secreted by obstinance and which bind to PS and link targets to receptors furthermore there's intracellular signaling pathways which is a very common pathway of receptor activation which uses the ceded 6 adapter or CD10 activation which leads to the cytoskeleton remodeling which then leads to phagocytic cup formation leading to internalization finally there's also a regulation of this selectivity which is the engulfment is selective and not random synaptic activity influences engulfment hence a weak or inactive synapses leads to signals that lead the glial to engulf it whereas active synapses express a the signal that lets people know or lets the glio know not to engulf it there are also lots of advanced imaging in quantification techniques such as super resolution microscopy electrophotography fluorescent receptors and genetic receptors in C elegans which allow us to view these functions in a very detailed manner such as the genetic reporters and C elegans which allows us to view trans genetic lines which express the glial specific molecules and neurons Pacific

| | |
|---|---|
| | molecules and allow us to visualize real time engulfment events in vivo this discovery of glial engulfment across species has shown microglia and mammals and shows how this engulf synapses are during post Natal development essentially in visual cortex furthermore another study has shown complement systems which mark less active synapses for microglial removal and the aristocrats show engulfing synapses debris using the mag F-10 and the ME RTK receptors the drosophilia glia in another study have shown Draper receptors and see these six adapters which Dr. engulfment of the synaptic terminal and the debris during metamorphosis and the glial pruning peripherals memory and synaptic remodeling pathways as well essentially this means that the way that draws aphelia glia encompasses synapsis during a metamorphosis kinda is similar to results in other mammals that also have this function which will be increasing sorry when studying and comparing glial usage and synapses between model organisms and humans furthermore there are lots of molecular cellular insights with signaling exposure recognition of the glial receptors internalization of the actin driven phagocytic cup and a degradation in lysosomes which is kind of the breakdown in like phagocytosis and key conserved pathways are the PS the complement C1Q or the C3 the Draper and the MERTK or M e.g. F-10 and this engulfment is an activity dependent and developmentally timed process which is ensuring that only the weak and excessive synapses are removed and not the ones that are necessary finally the relevance of diseases leads us to excessive engulfment which means linked to neurodegenerative disorders we can finally start finding a way to potentially get rid of this excess synapses and allow for the essential ones to survive more which could help with these neurodegenerative disorders in conclusion synaptic engulfment is not simply waste removal it is active regulated and instructive process that shapes the neural circuits through life and further research could show how glial engulfment interferes with neural activity and how glial involvement is essential for synapsis removal. |
| **Research Question/Problem/ Need** | The study investigates whether second-generation DNA methylation markers are associated with cognitive decline and neuropsychiatric symptoms in older adults. |

| | |
|---|---|
| **Important Figures** | Fig. 1.<br>  <br><br>Fig. 2: Spearman correlation between age and DNAm markers t baseline: DNAmPhenoAge, DNAmGrimAge, and DNAmTL |
| **VOCAB: (w/definition)** | DNA methylation: an epigenetic modification where mythl gorups are added to DNA – influences gene expression without changing the DNA sequence<br>Epigentic marker: Biochemcial feature such as DNA methylation that can indicate changes in gene regulation<br>Cognition: mental processes involved in knowledge, learning, memory,a dn problem-solving |
| **Cited references to follow up on** | Dayon L, Guiraud SP, Corthésy J, Da Silva L, Migliavacca E, Tautvydaitė D, Oikonomidi A, Moullet B, Henry H, Métairon S, Marquis J, Descombes P, Collino S, Martin FJ, Montoliu I, Kussmann M, Wojcik J, Bowman GL, Popp J (2017) *One-carbon metabolism, cognitive impairment and CSF measures of Alzheimer pathology: Homocysteine and beyond*. Alzheimers Res Ther 9, 43<br><br>Fortin JP, Triche TJ Jr., Hansen KD(2017) *Preprocessing, normalization and integration of the Illumina HumanMethylationEPIC array with minfi.* Bioinformatics 33, 558–560. |
| **Follow up Questions** | Which speceific DNA methylation markers would be most stringly associated with the cognitive decline?<br><br>What type of interventions could potentialy change the epigentic markers to reduce neuropsychiatric decline. |

# Article #16 Notes: Depression and inflammation: Correlation between changes in inflammatory markers with antidepressant response and long-term prognosis

| | |
|---|---|
| **Source Title** | Depression and inflammation: Correlation between changes in inflammatory markers with antidepressant response and long-term prognosis |
| **Source citation (APA Format)** | Kofod, J., Elfving, B., Nielsen, E. H., Mors, O., & Köhler-Forsberg, O. (2022). Depression and inflammation: *Correlation between changes in inflammatory markers with antidepressant response and long-term prognosis*. European Neuropsychopharmacology, 54, 116–125. https://doi.org/10.1016/j.euroneuro.2021.09.006 |
| **Original URL** | https://www.sciencedirect.com/science/article/pii/S0924977X21007562?via%3Dihub |
| **Source type** | Research Article |
| **Keywords** | Depression, inflammation, inflammatory markers, antidepressant response, prognosis cytokines, immune system |
| **#Tags** | Depression, inflammation, antidepressants, cytokines, prognosis |
| **Summary of key points + notes (include methodology)** | He begins by talking about how depression is a complex and often treatment resistant disorder frequently Co occurring with physical illnesses so great inflammation has been proposed as one of these biochemical markers that is linked to depression prior studies have shown an increased level of inflammatory markers such C-reactive protein (CRP) or interleukin-6(IL-6) and tumor necrosis factor-alpha (TNF-a) in MDD patients. However many early researchers have severe limitations such as most studies were cross sectional rather than longitudinal they measured few inflammatory markers at a single point, lacked to account for the heterogeneity of symptoms and few had long-term follow up data or controlled for BMI or smoking. Recent psychoimmuniology emphasizes multiple inflammatory amrkers over a course of time, symptoms-specific correlations (neurovegetative symptoms such as sleep, appetit and energy). The authors aimed to analyze a subgroup from the GENDEP rial to test whether inflammatory amrkers correlate with Baseline depression severity, specific symptom dimentions, short tern antidepressant response, and long term depression outcomes. They did this by using GENDEP trial (Genome based Theraptutic Drug and Depression) which is a European multicenter randomized study from 2004-2007 which focused on 90 Danush |

| | |
|---|---|
| | outpatients with moderate severe depression. The participants were 71% femail and had a mean ago of 38 years, administered with escitalopram (44% - anti depressant) or nortriptyline (56% - antidepressant) for 26 weeks. Used HAM-D (Hamilton), MADRS (Montgomery), and BDI (Beck) to measure depression. Symptom dimensions were derived statistically: mood, anxity, cognitive, neurovegetative, and suicidality. Inflammatory markers were 27 in tooal with pro and anti inflammatory cytokines and chemokines that were measurered periodically in 26 weeks using Bio-Plex multiplex assays. Further more two composite inflammation scores were createsd with Cytokine score (IL-1β, IL-6, IFN-γ, TNF-α), Chemokine score (IL-8, MCP-1, Eotaxin, RANTES, VEGF). Long term outcomes were linked with the Danish national registers with main outcomes from psychiatric hospital contacts. Statsictical anlysis involved the Pearson correlation (basline inflammatory markers vs symptom severity), Mixed-effect Models (logitutidnal changes in amrkers during treatment), and Cox regression (inflammatory markers vs. 10-year hospital contact risk). The article found that there was no correlation with overall depression severity and baseline inflammation however several markers (IL-5) correlated with specific symptom dementiosn such as a negative correlation with mood and anxiety and cognitive symptoms, whereas there was a positive correlation with suicidality and neurovegetative symptoms. IL-1β, IL-2, IL-4, IL-7, IL-8, IL-9, IL-13, IL-17, TNF-α and more inflammatory markers significantly decreased after/during the 26 weeks or treatment. Some weak associations emerged with changes in certain markers correlated with neurovegetative symptom improvement. Further more IL-9 and IL-12 changes differed moderately between the remitters and nonremitters. Furthermore the composite inflammation scores did not predict treatment repsonses. In conclusion inflammation and depression are linked in a symptom specific way but not a global way. |
| **Research Question/Problem/ Need** | The authors aimed to analyze a subgroup from the GENDEP rial to test whether inflammatory amrkers correlate with Baseline depression severity, specific symptom dimentions, short tern antidepressant response, and long term depression outcomes. |
| **Important Figures** |  <br>Fig 1. Correlation between baseline levels of the cytokine (A) and chemokine (B) composite inflammation markers with drugspecific |

| | |
|---|---|
| | treatment response after 12 weeks, measured as improvement on the MADRS (Kofod et al., 2022) |
| **VOCAB: (w/definition)** | Inflammatory markers: Biomolecules, often cytokines, that indicate activation of immune system<br>Cytokines: signaling protiens released by immune cells that mediate inflammation<br>Antidepressant response: improvement in depressive symptoms follwoing pharmacological treatment |
| **Cited references to follow up on** | MILANESCHI, Y., KAPPELMANN, N., YE, Z., LAMERS, F., MOSER, S., JONES, P.B., BURGESS, S., PENNINX, B.W.J.H. & KHANDAKER, G.M. 2021. *Association of Inflammation with Depression and Anxiety: evidence for Symptom-Specificity and Potential Causality from UK Biobank and NESDA Cohorts*.medRxiv,2021.01.08.20248710.<br><br>Uher, R., Farmer, A., Maier, W., Rietschel, M., Hauser, J., Marusic, A., Mors, O., Elkin, A., Williamson, R. J., Schmael, C., Henigsberg, N., Perez, J., Mendlewicz, J., Janzing, J. G. E., Zobel, A., Skibinska, M., Kozel, D., Stamp, A. S., Bajs, M., & Placentino, A. (2007). *Measuring depression: comparison and integration of three scales in the GENDEP study*. Psychological Medicine, 38(2), 289–300. https://doi.org/10.1017/s0033291707001730 |
| **Follow up Questions** | Which specific inflammatory markers showed the strongest correlation with antidepressant response?<br>Are there changes in inflammatory markers predicting relapse or long-term depression outcomes?<br>Are these findings generalized to broader populations with depression? |

# Article #17 Notes: Adolescent suicide assessment and management in primary care

| | |
|---|---|
| **Source Title** | Adolescent suicide assessment and management in primary care |
| **Source citation (APA Format)** | Aalsma, M., Keys, J., Ferrin, S., Shan, M., Garbuz, T., Scott, T., Adams, Z., Hulvershorn, L., & Downs, S. (2022). Adolescent suicide assessment and management in primary care. BMC Pediatrics, 22(1). https://doi.org/10.1186/s12887-022-03454-4 |
| **Original URL** | https://link.springer.com/article/10.1186/s12887-022-03454-4 |
| **Source type** | Research Article |
| **Keywords** | Adolescent suicide, primary care, assessment, risk management, mental health, screen, intervention |
| **#Tags** | Adolescent, suicide, primary care, screening |
| **Summary of key points + notes (include methodology)** | Suicide is a major public issue in the United states ranking the second leading cause of death among individuals 10 to 14 in 2020 approximately 18% of adolescents between the ages 13 to 18 report having suicidal ideations with significant increase of their risk of attempting suicide alarmingly more than 80% of adolescents who attempt suicide have already received some sort of mental health care many primary care settings offer key opportunities for prevention because of over half of adolescent visits to a primary care provider annually and many individuals who die from suicide have visited a health care provider within weeks of their death however standardized management and guidelines for following up for suicide risk in adolescents are lacking existing frameworks such as the glad PC the guidelines for adolescent depression in primary care and suicide prevention Resource Center recommend initial safety planning but these resources do not help manage beyond ongoing follow-ups and other events that are not taken into account prior studies in the UK and the US have shown that there have been very limited mental health follow-ups after self harm incidents and lots of inconsistent referral practices with the UK having 55% of the youth with several harm history had no recorded referral within 12 months given this gap the current study aimed to describe how suicide management and follow up can occur in a primary care setting focusing on adolescents who screened positive for suicide risk the study was designed as a descriptive retrospective chart review of 200 adolescents between the ages 12 to 20 years old who were screened positive for suicide risk in the Midwestern United States primary care clinics between the years 2014 and 2017 they used the child health improvement through computer |

| | |
|---|---|
| | automization chica system a computer decision support system CSS that use a pre Screener form of PSF and the pre Screener question was have you ever seriously thought about killing yourself data plan or actually try to kill yourself response is automatically generated A physician worksheet prioritizing the youths health and recommended actions as per the bright futures guideline the participants had a mean age of 14.7 years with a standard deviation of two the sax was 27% female and raised was 65% black clinics were urban and affiliated with local county hospitals and were staffed by Pediatrics and adolescents medicine specialists researchers reviewed medical notes for identified patients across visits during the study window there are extracted included demographics such as their age race and sex their suicide history ideation attempts and any thoughts or ideas or any actions that they've committed physician actions such as safety plans or referrals behavioral health referrals and visits and psychiatric medication and prescriptions and follow-ups they used descriptive statistics and univariate comparisons using the T test the wilcon or the chi square test they also used categories of suicide risks such as suicidal concern or no concern and analysis performed within SAS of 9.4 was significant threshold of P less than 0.05 this article sums up the results with finding out of 200 adolescents endorsing suicide risk with 39 of them which is 90 point 13.5% were considered suicidal concert by physicians active suicidal ideation and past attempts were very high among the suicidal concerned group with a value of less than 0.001 and past ideations 67% were concerned whereas 38% were no concern group and the depression diagnosis 72% of the group was a concern whereas 52% was no concern and behavioral health referrals 72% were concerned and 46 were no concern safety environment assessment were rare only 13% were asked about weapons at home and only 7% were asked about having a safety plan psychiatric medication initiation was only 35% overall and then after the follow within 12 months span 141 adolescents or 70.5% had at least one follow up visit mental health statuses were discussed in 80% of these visits there was no significant differences between the groups and suicidal ideation or attempts at the follow up the behavioral health referrals the inquiry about weapons and the inquiry about safety plans however 15% of the patients were hospitalized with no group differences and then the continuity of medication use was slower and this led to an overall conciseness that physician practices need to be fixed with primary care providers showing inconsistent adherence to suicide management best practices as there have been low rates of firearm safety discussions and higher rates of behavioral health referrals possibly due to a fear of malpractice moreover medication continuity of among adolescents is very limited echoing prior research citing barriers like forgetfulness side effects difficulty filling prescriptions lack of structured medication plans furthermore there is a need for primary |

| | |
|---|---|
| | care based guidelines because although frameworks like glad PC provide guidance for depression and initial suicide screening clear evidence based follow-up protocols are missing and so individuals with suicide are often not able to be diagnosed or helped properly due to the lack of agility within these guidelines. |
| **Research Question/Problem/ Need** | The study examines how primary care providers assess and manage suicide risk in adolescents, aiming to identify the best practices, gaps, and challenges in suicide prevention in primary care settings. |
| **Important Figures** | **Table 1** Variables from medical record<br>**Variable Label** / **Description** / **Response Scale**<br>**Basic Visit Information**<br>Sex — Sex of the patient — Numeric score<br>Age at incident visit — Provider reported the age of the patient at the visit — Free Response<br>Race — Provider reported the race of the patient at the visit — Free Response<br>**Suicidal Ideation/Attempts & Self-Harm**<br>Active suicide ideation — Provider reported if patient had active suicidal ideation — Yes/No<br>Past suicide ideation — Provider reported if patient had a history of suicidal ideation — Yes/No<br>Past suicide attempt(s) — Provider reported if patient had past suicide attempt(s) — Yes/No<br>Suicide attempt history — Provider reported details regarding patient's past suicide attempt(s) — Free Response<br>Weapons — Provider reported asking if patient had any weapons at home — Yes/No<br>Safety plan — Provider reported asking patient about safety plan — Yes/No<br>**Behavioral Health**<br>Mental health referral — Provider referred patient to outpatient mental health services — Numeric score<br>Co-morbid diagnoses — Behavioral health-related diagnoses listed by the provider — Free Response<br>Hospitalizations[a] — Provider reported if patient had been in an inpatient psychiatric setting since previous visit — Numeric score<br>Visit engagement[a] — Provider reported if patient has attended behavioral health visits — Numeric score<br>**Psychiatric Medication**<br>Psychiatric medications — Provider reported starting patient on psychiatric medication — Yes/No<br>Psychiatric medication list — List of psychiatric medications prescribed to patient — Free response<br>Psychiatric medication continuity[a] — Provider reported if patient continued to take prescribed psych meds — Numeric score<br>[a] Coded only at follow-up visit<br>Table 1. Demographic and clinical characteristics of adolescents included in the suicide assessment in study |
| **VOCAB: (w/definition)** | Suicide assessment: evaluation of indivual's risk of attempting suicide, typically through interviews and screening tools<br>Risk management: stratergies to preent or mitigate adverse outcomes, here referring to interventions for suicide prevention |
| **Cited references to follow up on** | Dueweke AR, Bridges AJ. *Suicide interventions in primary care: A selective review of the evidence*. Fam Syst Health. 2018;36(3):1–15<br><br>Gould MS, Greenberg T, Velting DM, Shaffer D. *Youth suicide risk and preventive interventions: A review of the past 10 years*. J Am Acad Child Adolesc Psychiatry. 2003;42(4):386–405. |
| **Follow up Questions** | Which assessment tools are most effective for identifying suicide risk in adolescents in primary care?<br><br>Which charecteritics are focused on inorder to assess adolescent suicidal ideation? |

# Article #18 Notes: Serotonin transporter: a potential substrate in the biology of suicide

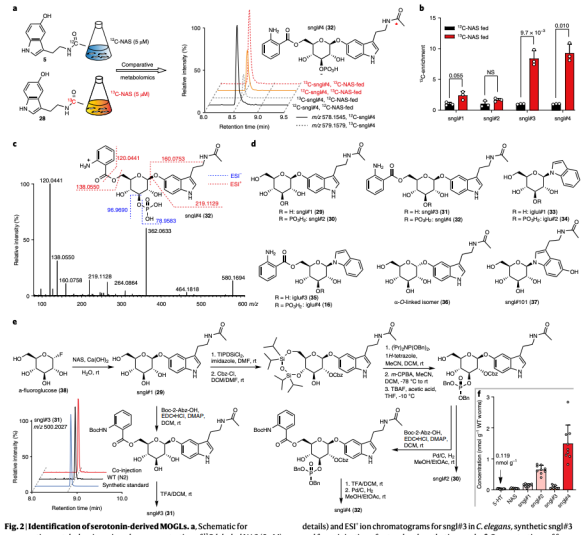
| | |
|---|---|
| **Source Title** | Serotonin transporter: a potential substrate in the biology of suicide |
| **Source citation (APA Format)** | Purselle, D. C., & Nemeroff, C. B. (2002). *Serotonin Transporter: A Potential Substrate in the Biology of Suicide*. Neuropsychopharmacology, 28(4), 613–619. https://doi.org/10.1038/sj.npp.1300092 |
| **Original URL** | https://www.nature.com/articles/1300092 |
| **Source type** | Review Article |
| **Keywords** | suicide; serotonin transporter; post mortem; platelets; genetics; neurobiology |
| **#Tags** | Suicide, neurobiology, genetics |
| **Summary of key points + notes (include methodology)** | Suicide is a increasingly serious public health problem in the US yet its nerobiological underpinning poorly understood. Suicide is highly cooreralted with depressive symptoms, and considerable evidenvece siggest that depression is associated with a relative deficiency in serotonergic neurotransmission. The serotonin circuits also mediate impulsive very which is a trait that is very relevant to suicide these findings one put together suggests that alternations in the serotonergic system might contribute to suicidal behavior serving as a key signal for researchers to scrutinize the serotonin transporter as ER team as a potential substrate for the biology of suicide in the article they use postmortem brain tissue platelets and DNA from suicide completers in attempters to find evidence for a preeminent role for of SRT in the pathology of suicide. Suicide has become an increasingly common health issue concerning a very large part of the population in the world with suicide being the 8th leading cause of death for all citizens and the third leading cause of death for those between the ages of 15 and 24 risk factors for suicide include the presence of one or more psychiatric illnesses such as mood anxiety disorders bipolar disease panic disorders and many more other long term risk factors are single marital status living alone unemployment and other external factors so investigations of biological substances of suicide risk were issued almost 30 years ago with the first evidence of serotonergic system alterations in the brain of suicide emerging in 1976 with asberg at all which demonstrated decreased levels of 5-HIAA in the CSF for suicide attempters. Evidence also suggests that the serotonergic system is altered in suicidal behavior has promoted investigations to scrutinize the role that SRT in plays in the pathophysiology of suicide the Sir is believed to be primarily responsible for |

| | |
|---|---|
| | the termination of action ohh 5 HD after it is released from the nerve terminal into the synapsis over the last 20 years several studies have appeared in which the alternation of SERT in its binding and postmortem brain tissue of suicide victims were measured and compared in various control groups the early investigation of Sarah binding and suicide victims was more recent ones used HM neoprene as the ligand and desperation as a replacing agent to define specific binding in the brain at least 26 studies of postpartum brain SCRT binding and suicide victims has been completed yielding very inconsistent data with the frontal cortex has been the most widely studied brain region while numerous brain regions have received less attention. However the prefrontal cortex of hypothalamus have also showed to be relevant in the decreased SRT density but there might be differences that also exist between violent and nonviolent suicide the some studies observed decrease in the frontal escrt binding others increased and many did not change Lawrence at all in 1998 found that nonviolent suicide showed decreased SRT levels in the putamen or the hippocampus possibly reflecting on the HP a access damage from poisoning and so the authors attribute the lack of consciousness to several methodological confounds including ligand specialties such as H-imipramine Which binds to multiple receptor sites beyond the ERT another methodological confound includes the tissue preparation which is when homogeneous tissue methods can lose this specificity of region and may be affected by the unprecise separation of grey and white matter another mythological confound would be antidepressant treatments where many studies fail to exclude subjects with premortem psychotropic use which also alters the SRT binding and finally variability factors such as age sex diagnostic post board of intervals shortage of time can also lead to this unclarity the art in platelets is also very important because researchers use platelets as peripheral models for studying teratogenic functions since platelets contain SRT and five HT2A which are similar to those in the CNS numerous studies have examined the platelets are density and depressed or suicidal individuals the third gene polymerase it is also related to the chromosome 17 Q 11.1 and 17 Q 12 which span around 31 kilobase pairs and contain 14 exons and they have two main allies along in a short version the long allele exhibits higher transcription activity and greater serotonin uptake whereas a short allele is linked to reduction transporter expression although SER T genotype influences the expression and virtue its relationship to suicide behavior has been inconsistent across studies approximately 14 genetic association studies have tested the link between SRT and suicide with half finding no association and half reporting some association so overall the authors conclude that assert transporters likely play a role in the biology of suicide the exact magnitude and nature still remains to be unclear however and the neurochemical abnormality and suicides are not limited to one protein |

| | |
|---|---|
| | or transmitter in the alteration insert must be understood within the broader context of suicide and the reduced serotonergic activity the authors hoped to find that fine functional brain imaging techniques such as a PET or an SPECT Which would play an increasingly important role in identifying receptors and transporters alteration and living subjects furthermore researchers hope to explore signal transduction pathways gene expression pathways and molecular mechanisms. |
| **Research Question/Problem/ Need** | THe article reviews the evidence that alteration in the SERT transporter system may contribute to the biological mechanisms that underlie suicide |
| **Important Figures** | **Table I** Serotonin Transporter Binding Studies in Post-mortem Brain Tissue of Suicide Victims<br>**Region showing change in suicide victims** / **Method** / **Ligand**<br>*Increases in SERT binding*<br>Meyerson *et al* (1982) ↑ Frontal cortex Homogenate [$^3$H]-IMI<br>Gross-Isseroff *et al* (1989) ↑ Hippocampus Autoradiography [$^3$H]-IMI<br>Arato *et al* (1991) ↑ Left frontal cortex Homogenate [$^3$H]-IMI<br>*Decreases in SERT binding*<br>Stanley *et al* (1982) ↓ Frontal cortex Homogenate [$^3$H]-IMI<br>Paul *et al* (1984) ↓ Hypothalamus Homogenate [$^3$H]-IMI<br>Crow *et al* (1984) ↓ Frontal cortex Homogenate [$^3$H]-IMI<br>Gross-Isseroff *et al* (1989) ↓ Postcental gyrus, insula, claustrum Autoradiography [$^3$H]-IMI<br>Lawrence *et al* (1990a) ↓ Putamen Homogenate [$^3$H]-parox<br>Arato *et al* (1991) ↓ Right frontal cortex Homogenate [$^3$H]-IMI<br>Laruelle *et al* (1993) ↓ Frontal cortex Homogenate [$^3$H]-parox<br>Arango *et al* (1995) ↓ Prefrontal cortex Autoradiography [$^3$H]CN-IMI<br>Dean *et al* (1996) ↓ Hippocampus Homogenate [$^3$H]-parox<br>Lawrence *et al* (1997) ↓ Putamen Homogenate [$^3$H]-parox<br>Rosel *et al* (1997) ↓ Hippocampus Homogenate [$^3$H]-IMI<br>Rosel *et al* (1998) ↓ Hippocampus Homogenate [$^3$H]-IMI<br>Lawrence *et al* (1998) ↓ Putamen Homogenate [$^3$H]-IMI<br>Mann *et al* (2000) ↓ Ventral prefrontal cortex Autoradiography [$^3$H]CN-IMI<br>*No change in SERT binding*<br>Owen *et al* (1986) None Homogenate [$^3$H]-IMI<br>Arora and Meltzer (1989) None Homogenate [$^3$H]-IMI<br>Lawrence *et al* (1990b) None Homogenate [$^3$H]-parox<br>Arora and Meltzer (1991) None Homogenate [$^3$H]-IMI<br>Hrdina *et al* (1993) None Homogenate [$^3$H]-parox<br>Mann *et al* (1996) None Homogenate [$^{33}$H]-parox<br>Little *et al* (1997) None Autoradiography [$^{125}$I]-RTI-55<br>Rosel *et al* (1997) None Homogenate [$^3$H]-parox<br>Rosel *et al* (1998) None Homogenate [$^3$H]-parox<br>Du *et al* (1999) None Homogenate [$^3$H]-parox<br>Bligh-Glover *et al* (2000) None Autoradiography [$^3$H]-parox<br>Arango *et al* (2001) None Autoradiography [$^3$H]CN-IMI<br>IMI, imipramine; parox, paroxetine; CN-IMI, cyanoimipramine.<br>Table 1. summary of serotonin transporter studies related to suicide (with specific brain area and molecule – based on other articles) |
| **VOCAB: (w/definition)** | Serotonin transporter (5-HTT): protein responsible for reuptake of serotonin from the synaptic cleft, regulating serotonergic signaling<br>Neurotransmission: the process of communication between neurons via chemical signals<br>Depression: MDD charecterized by perisistent sadness, with sucicidal behavior falling under MDD |
| **Cited references to follow up on** | Lawrence KM, De Paermentier F, Cheetham SC, Crompton MR, Katona CL, Horton RW (1990a). *Brain 5-HT uptake sites, labelled with [3H]paroxetine, in antidepressant-free depressed suicides*. Brain Res 526: 17–22. |
| **Follow up Questions** | How do genetic polymorphism in the serotonin transporter affect suicide risk?<br>What might antidepressants targeting 5-HTT influence suicide risk?<br>Are there specific biomarkers? |

Article #19 Notes: Parallel pathways for serotonin biosynthesis and metabolism in C. elegans

| | |
|---|---|
| **Source Title** | Parallel pathways for serotonin biosynthesis and metabolism in C. elegans |
| **Source citation (APA Format)** | Yu, J., Vogt, M. C., Fox, B. L., Wrobel, C. J. J., Diana Fajardo Palomino, Curtis, B. J., Zhang, B., Le, H. H., Arnaud Tauffenberger, Hobert, O., & Schroeder, F. C. (2022). Parallel pathways for serotonin biosynthesis and metabolism in C. elegans. 19(2), 141–150. https://doi.org/10.1038/s41589-022-01148-7 |
| **Original URL** | https://pubmed.ncbi.nlm.nih.gov/36216995/ |
| **Source type** | Research article |
| **Keywords** | Serotonin, biosynthesis, metabolism, *C. Elegans,* neurotransmitters, enzymatic pathways |
| **#Tags** | Serotonin, C. elegans, biosynthesis, neurochemistry |
| **Summary of key points + notes (include methodology)** | Serotonin (5-HT) is a increasingly conserved neurotransmitter which regulates the body's behavior and physiological interactions across a wide range of species, with specific behaviors including locomotion, feeding, egg-laying, and mood-regulation. In C. Elegans, the serotonin biosynthesis pathway was traditionally thought to happen in the serotonergic neurons through the TPH-1 hydroxylase pathway, however new observations show that tph-1 mutants retain serotonin immunoreactivity and serotonin dependent behaviors which suggest there may be additional biosynthesis pathways. The writers of the article discovered the PAH-1 which acts as a second enzyme which is capable of hydrolyzing the tryptophan, operating in parallel with the TPH-1 which originally hydrolases the tryptophan. The genetic analyses show:<br>1. Bas-1 null mutant had no serotonin or serotonin-derived metabolites which confirmed BAS-1 as the only decarboxylase<br>2. Tph-1 mutant had 20-50% of serotonin metabolites<br>3. Pah-1 mutant showed a huge reduction in serotonin derivatives<br>4. Tph-1 + pah-1 double mutant shows a complete abolishment in serotonin metabolite production; hence these two combined molecules contribute to the serotonin biosynthesis with pah-1 playing a huge role in non-neuronal tissue.<br>The PAH-1 expession is localized to epidermal cells, seam cells, and tail |

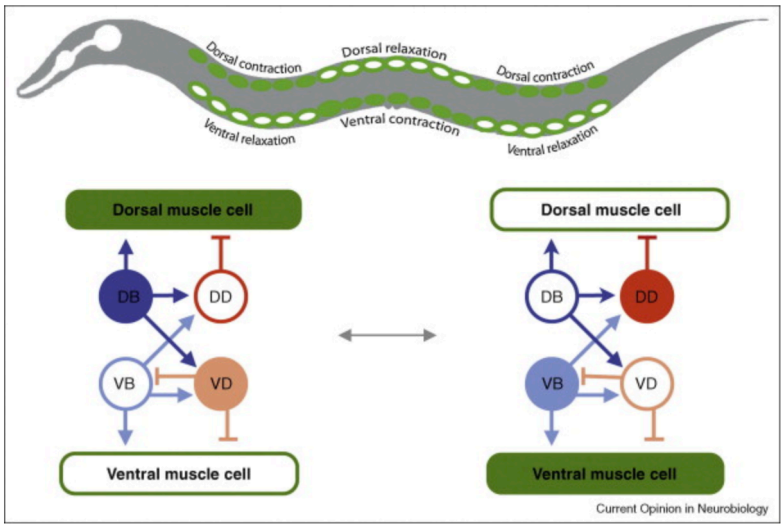
| | |
|---|---|
| | socket glial cells. Additonally the endogenous tagging shows that BAS-1 is expressed in the epidermis and intestine which overlaps witht he PAH-1. The co-expression of these two molecules allows for support in the cell-autonomous serotonin synthesis outside the neurons. The free serotonin represents only a small part of the total serotonin-related meotobnilites beaucase most serotonin in rapidly convered into NAS which is then converted to a glucosylation leading to phosphorlaytion and anthranilate modification. The CEST-4 is expressed in the intensie and epidermis and is required for the biosynthesis for the reuptake inhibitor to function. Additonaly the loss of PAH-1 leades to increased exploratory behvior similar tot he tph-1 mutant and reduces egg-laying responses to antidepressants such as fluoxzetine. The lack of the NAS supplement is particially helpful in rescuing the exploraroty hbehvaior in Serotonin-deficent bas-1 mutants whcih show that serotonin metobiloties (NAS and sngls) actively regulate behvaior in additon to free serotonin. Overall this study shows that the srotonin biosynthesis and metoablism in C. elegans is more complex hat previsously thought out sysnthesis pathways, whsoing the exsiitwence of a parallel non-neuronal pathways that shows biologically active serotonin-derived molecules that contribute to siganling and bahvior in an organism. |
| **Research Question/Problem/ Need** | The paper investicgates whether serotonin in C. elegans is synthesized exculsively in the TPH-1 pathwyas or whethere there is an alternative, non-neuronal biosytheethuc and metaboloic pathway that contributes to serotonin prodcution and function. |
| **Important Figures** |  <br>**Fig. 2 \| Identification of serotonin-derived MOGLs. a**, Schematic for comparative metabolomics using a low concentration of $^{13}$C-labeled NAS (5 µM). HPLC traces demonstrate $^{13}$C-labeling of new serotonin metabolite, sngl#4. **b**, $^{13}$C-enrichment of the four most abundant NAS-derived metabolites. **c**, ESI$^+$ MS/MS spectrum and proposed fragmentation of sngl#4. **d**, Structures of identified serotonin-derived MOGLs and related MOGLs incorporating indole instead of serotonin. **e**, Synthetic scheme for sngl#1–4 (see Supplementary Information for details) and ESI$^+$ ion chromatograms for sngl#3 in *C. elegans*, synthetic sngl#3 and for coinjection of natural and synthetic sample. **f**, Concentrations of free serotonin (5-HT), NAS, sngl#1, sngl#2, sngl#3 and sngl#4 in WT *C. elegans*. Data in **b** ($n = 3$) and **f** ($n = 9$, except for 5-HT measurement, where $n = 12$) represent biologically independent experiments, and bars indicate mean ± s.d., *P* values calculated by unpaired, two-tailed *t*-test with Welch correction; NS, not significant; rt, room temperature.<br>Fig 2. Genetic and biochemical analysis of enzymes involved in serotonin synthesis |

| | |
|---|---|
| **VOCAB: (w/definition)** | Serotonin: a neurotransmitter involved in mood, appetite, sleep and other physiological functions<br>Biosynthesis: the enzymatic production of compelx molecules from simpler precursors<br>Metabolism: the chemical processes that modify or break down molecules within an organism |
| **Cited references to follow up on** | Livak, K. J. & Schmittgen, T. D. *Analysis of relative gene expression data using real-time quantitative PCR and the 2(T)(-Delta Delta C) method*. Methods 25, 402–408 (2001). [use to learn about PCR test – might need for genetic analysis?] |
| **Follow up Questions** | How dot he parallel sertonin pathways differ in their regulationa nd fucntional output?<br>How does serotonin metablism interact with other neurotransmitters in C. elegans? |

Article #20 Notes: Caenorhabditis elegans: a model system for systems neuroscience

| | |
|---|---|
| **Source Title** | Caenorhabditis elegans: a model system for systems neuroscience |
| **Source citation (APA Format)** | Sengupta, P., & Samuel, A. D. T. (2009). C. elegans: a model system for systems neuroscience. *Current Opinion in Neurobiology*, *19*(6), 637–643. https://doi.org/10.1016/j.conb.2009.09.009 |
| **Original URL** | https://www.sciencedirect.com/science/article/abs/pii/S0959438809001330?via%3Dihub |
| **Source type** | Review Article |
| **Keywords** | C. elegans, systems neuroscience, neural circuits, behavior, model organism, neurobiology |
| **#Tags** | C. elegans, neuroscience, neural circuits |
| **Summary of key points + notes (include methodology)** | The article reviews hot C. elegans enable systems neuroscience, which is linked to molecular and cellular mechanisms to matching behavioral assays through complete or near-complete knowledge. Through top-down |

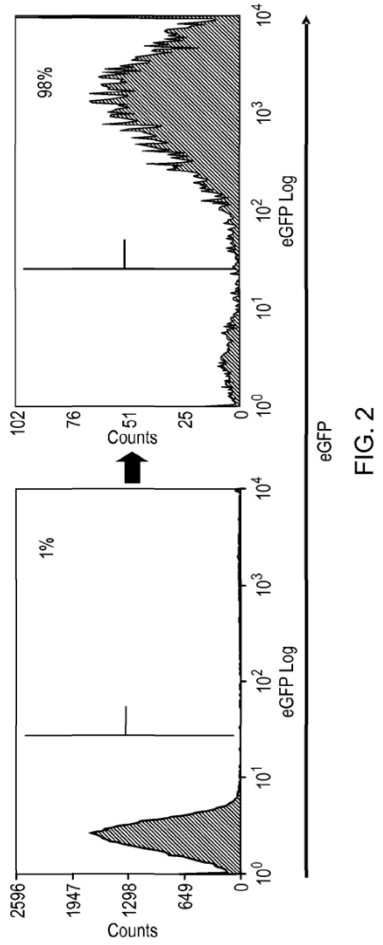
| | |
|---|---|
| | integreation such as quanatative behavioral analysis, and bottom-up integratoon like genetics and synaptic control, the authors were able to determine the accuracy of using C. elegans as a model for systems of neuroscience. The study looks at the anatomical strucure though the electron mucroscope which procdes a compeletely wired diagram of the worm's nervous system. Additionally, reconstructio helps fill the gaos un the ventral nerve cord motor which completes the synaptic maps for 58 motor neurons. The behavioral qualities of locomotion include controlled mechanical environments, with high speed tracking videos to extract waveform parameters such as frequency, amplitude, and wavelength. Additonally the calcium imaging (genetically encoded calcium indicators were expressed) showed muscle cells, motor neurons, and command interneurons. Additionally optogenentic techniques were used to selectively activate or stop specific neurons and muscle cells which allows actual testing of indivual circuit parts that contribute to locomotion and behavior. The electrophysiology recordings combined with the optogenetic stimulation show the synaptic transmission in C. elegans is greaded rather than all or nothing which supports the continous modulation of motor outputs. Overall the integration os a compelte circuit anatomy with rpecise genetics and optical manipulation, high resultion behavior tracking and quanittative computairtional analysis show that C. elegans provide a new powerful and experiemntally tractable model for systems neuroscince. Through direct enabling between molecular and cellular mechanisms, ciruit dynamics, and measurable behavior in a living organism, this work shows how complex neural computations can be understood across multiple biological experiments. |
| **Research Question/Problem/ Need** | How can the complete or near complete knowledge of neural circuitry in C. elegans combined with quotative behavioral analysis and targeted neural manipulation be used to understand the sensory inputs and how they are transformed into the motor behaviors at the stems level. |

| | |
|---|---|
| **Important Figures** |  <br>Fig 1. Layout of C. elegans nervous system, highlighting sensory, motor and interneurons. |
| **VOCAB: (w/definition)** | Systems neuroscience: feild of neurosceince focused on how networks of neurons interact to produce behaviors and process information<br>Neural circuits: networks of interconnected neurons that process information and control behaviors<br>Sensory processing: the mechansim by which organisms detect and resoind to external stimuli |
| **Cited references to follow up on** | Srivastava, N., Clark, D. A., & Samuel, A. D. T. (2009). Temporal Analysis of Stochastic Turning Behavior of Swimming C. elegans. *Journal of Neurophysiology*, *102*(2), 1172–1179. https://doi.org/10.1152/jn.90952.2008<br><br>Wakabayashi, T., Kitagawa, I., & Shingai, R. (2004). *Neurons regulating the duration of forward locomotion in Caenorhabditis elegans*. Neuroscience Research, 50(1), 103–111. https://doi.org/10.1016/j.neures.2004.06.005 |
| **Follow up Questions** | What makes C. elegans uniquely suited for systems-levelstudies compared to more complex organisms?<br>How can genetic tools advance systems neuroscience research in this model? |

Patent #1 Notes: Marker–Suicide Gene Useful in Adoptive Cell Therapy

| | |
|---|---|
| **Source Title** | Marker–Suicide Gene Useful in Adoptive Cell Therapy |
| **Source citation (APA Format)** | Pulé, M., & Philip, B. (2021, February 23). *Marker–suicide gene useful in adoptive cell therapy* (U.S. Patent No. 10,925,943 B2). U.S. Patent and Trademark Office. https://patents.google.com/patent/US10925943B2 |
| **Original URL** | https://patents.google.com/patent/US10925943B2 |
| **Source type** | Patent |
| **Keywords** | Marker-suicide gene, adoptive cell therapy, CAR-T cells, safety switch, gene engineering, immunotherapy |
| **#Tags** | AdoptiveCellTherapy, CAR-T, SucideGene |
| **Summary of key points + notes (include methodology)** | The patent is about a new engineered polypeptide which is designed for adoptive cell therapy which is a form of immunotherapy where genetically modified immune cells are infused into patients to fight caner or infection. The invention provides a compact dual-function, a “marker-suicde” gene which leads to selection and tracking of genetically modified cells and safe elimination of those cells (“suicide”). The double function design allows for safety, control, efficiency of the ACT such as CAR-T or TCR-T cell therapies. Adoptive cell theapies use modified T-cells to target cancers or infections (lymphoma, melanoma, etc.). However although ACT is effective it has serious risks including cytokine storms, autoimmunity, graft-vs-host diseases and vector insertion mutagenesis. To mititgate these risks they use a suicide gene to selectively destroy engineered cells when needed using exsisiting suicide systems such as HSV-TK or iCasp9. However they also have limitations such as HSV-TK is highly immunogenic and block antiviral drug use hwereas iCasp9 requires an experimental small molecule not widely available. There is a big need for ACT to track and purify tranduced T cells. Furthermore the exsisting marker genes are large/biologically active which is a safety hazard. The invention – Compact Marker Suicide Polypeptide- provides a chimeric polypeptide that combnes a marker epitope from CD34 (which recognized the clinical antibody QBEnd10), and the suicide epitope from CD20 (recognized by the monoclonical antibody Rituximab), which allows cell selection and controlled cell deletion. The protein’s overall formula: $St – R_1 – S_1 – Q – S_2 – R_2$ |

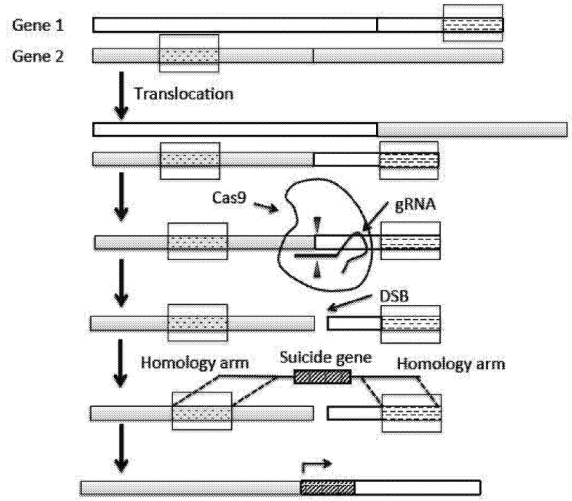
| | |
|---|---|
| | St = stalk sequence (projects the epitopes from cell surface)<br>$R_1$/ $R_2$ = Rituximab-binding epitopes<br>Q = QBEnd10-binding CD34 epitope<br>$S_1$/ $S_2$ = Optional spacer sequence (glycine/serine links)<br>The invention RQR8 gene encodes a 136 amino acid fusion protein when expressed in T cells the QBEnd10 allows selection and monitoring whereas the Rituximab administers the elimination of these cells if toxicity occurs. There were five experimental tests run. 1. Mapping the Marker Epitope which identified a 16 amino acid sequence on the CD34 which allows efficient magnetic sorting of transduced T cells. 2. Spacer Optimization which added the CD8-derived stalk and the optional linker to elevate the marker for the antibody better, resulting in the binding equivalent to full length CD34. 3. Combining with Rituximab Epitopes which tested multiple CD20-derived mimetopes and selected versions that maintained strong Rituximab binding and effective complement-mediated killing. 4. Compact Integration, which created compact construction (RQR8) with both CD34 and CD20, and functioned effectively for both sorting and cell ablation. 5. Vivo testing used in mouse GvHD model where T cells express RQR8 could be selectively tracked and removed using Rituximab which prevent severe immune complications. Applications of this invention include Adoptive Cell Therapy (ACT), Clinical Manufacturing (Easier purification using exsisiting QBEnd10), Safety Management ( immediate elimination of transduced cells), and Research/Monitoring (through vivo tracking and controlled deletion of modified cells). In conclusion the invention allows for a safety and control switch for immune cells which allows researchers to select and purify modified therapeutic cells using CD34 epitope detection, track the cells inside the body, and destroy the cells on demand with Rituximab if side effects occur, which greatly improves clinical safety and manufacturing simplicity of CAR-T and TCR-based cancer immunotherapy. |
| **Research Question/Problem/ Need** | Adresses the need for a controllable safety mechanism in adoptive cell therapies such as Car-T treatments by incorportating a marker-lines suicide gene. [could be used in my experiment if I found a lab to kill SERT reuptake] |

| | |
|---|---|
| **Important Figures** |  <br>Fig 2. functional overview of inducible cell elimination through the use of suicide gene |
| **VOCAB: (w/definition)** | Sucide Gene: a agenetic element that can trigger cell death when activated<br>Marker gene: a gene used to identify or select genetically modified cells,<br>Adoptive cell therapy: a treatment involving infusion of modified immune cells to target diseases such as cancer<br>CAR-T cells: chimeric antigen receptor T cells engineered to recognize and kill specific cancer cells<br>Safety Switch: a controllable mechanism to eliminate therapeutic cells to prevent adverse effects. |
| **Cited references to follow up on** | Lamers, C. H. J., Sleijfer, S., Vulto, A., Kruit, W. H. J., Kliffen, M., Debets, R., Gratama, J. W., Stoter, G., & Oosterwijk, E. (2006). Treatment of metastatic renal cell carcinoma with autologous T-lymphocytes genetically retargeted against carbonic anhydrase IX: First clinical experience. *Journal of Clinical Oncology,* 24(13), e20–e22. https://doi.org/10.1200/JCO.2005.04.5245 |

| | |
|---|---|
| **Follow up Questions** | How is the marker-suicide gene activated to trigger cell death?<br>What advantages does this system have over previous safety switches like iCasp9?<br>Can this be made to apply to multiple types of cell therapies? |

Patent #2 Notes: Methods and Compositions for Selectively Eliminating Cells of Interest

| | |
|---|---|
| **Source Title** | Methods and Compositions for Selectively Eliminating Cells of Interest |
| **Source citation (APA Format)** | Zhu, J., & Zhang, Y. (2020). Methods and compositions for selectively eliminating cells of interest (European Patent No. EP 3230460 B1). European Patent Office. https://patents.google.com/patent/EP3230460B1 |
| **Original URL** | https://patents.google.com/patent/EP3230460B1 |
| **Source type** | Patent |
| **Keywords** | Cell elemination, suicide gene, selective cell targeting, gene therapy |
| **#Tags** | Cell Therapy, sucidal gene |
| **Summary of key points + notes (include methodology)** | The patent introduces a new genetic engineering approach to design a selectively elimination processes for specific cells in cancer while leaving healthy cells unharmed. The project's motivation is derived from the limitations in traditional cancer treaments which lack the selectivity and casue severe side effects by damaging normal tissue. Earlier methods use the suicide gene of CRISPR but none of them effectively combine precise DNA targeting with self destructive emchanisms that only activate the diseased cells. The investion is centered around the CRISPR/Cas-based genetic system that introduced a sucide gene into the cell that contrains a unique genetics marker (chromosomal translocation in cancer cell). Once the gene is integrated the suicide gene truggers the cell to self destruct either naturally or upon exposure to a specific compound. The key components include a guide RNA (gRNA), which is designed to hybridize a taret DNA sequence unique to the cell of intrest, targeting breaking point jnctions created by chromosomal translaocation). The cas Protien (Cas9) acts as the molecular scissor that create the double strand breaks at precise target sites, can be wild type (cuts both DNA strands) or modified. The suicide gene  a gene when expressed causes cells to die. Vectors which carry DNA (viral and non viral) contain sequences encoding the gRNA, Cas Protein, and suicide gene. Finally the promoters control the expression of Cas9 or the suicide gene, with cancer-specific promoters (telomerase, PSA, CEA) which ensure the gene activation. The engineered vector is delivered to the cells (via lipofection, electroporation, or viral infection), then the CRISPR/Cas9 ssytem regonizes and binds to a specifc C=cancer DNA sequences. Here the Cas performces a double strand break and the suicide |

| | |
|---|---|
| | gene integrates into the genome at this sit. Once it is integrated the suicde gene is expressed, leading to the cell death, with the healthy cells remaining unaffected since they lack the unique target sequence. Cancer specific applications (in chromosal rearrangements) include Chronic Myelogenous Leukemia (CML), Burkitt's Lymphoma, Mantle Cell Lymphoma, and Follicular Thyroid Cancer. The patent had a modeled experiment where they used human cells and targeted the AAVS1 locus on the 19th chromosome and inseted the HSV-TK suicde gene using contructied palsmids that express Cas9 and gRNA (PX330 plasmid). They introduced the plasmid to the sucide gene and integrated it into the human cell to trigger apoptosis through GCV treatment. This treatment allows high specificity, reduced side effects, is versatile, is adaptable, and is personalized medication based on tumor genetics. Overall the patent presents and innovative CRISPR based suicde gene therpy system capable of targeting and eliminating specific cells with unmatched precision through the merging of genome-editing accuracy with controlled cell self-destruction. |
| **Research Question/Problem/ Need** | The need for a method to selectively eliminate a specific cell population to enhance safety and control in gene |
| **Important Figures** |  <br>FIG. 1<br>Fig 1. A overall view of selective cell elimination system |
| **VOCAB: (w/definition)** | Suicide gene: genetic element that can induce cell death when activated<br>Selective elimination: targeted removal of specific cell population without affecting others<br>Gene therapy: the use of genetic material to treat or prevent disease |

| | |
|---|---|
| **Cited references to follow up on** | Li, W., et al. (2013). *Safeguarding clinical translation of pluripotent stem cells with suicide genes*. Organogenesis, 9(1), 34–39. https://doi.org/10.4161/org.24317 |
| **Follow up Questions** | How can the inducible suicide gene activated in such a way that it only eliminates the target cells?<br>Can this be applied to the SERT gene? |