

Effectiveness of Normalising Neurotrophic Signaling as Treatment Strategies for Huntington's Disease

Zita Ng¹ and Kenneth Kuhn[#]

¹ Horizon Academic, USA

[#]Advisor

ABSTRACT

Huntington's Disease (HD) is an inherited neurodegenerative disorder. It is established that BDNF deficiency and the imbalance of the p75NTR/TrkB expression are responsible for striatal atrophy in HD patients, which collectively provide a molecular explanation to the most significant hallmark symptom of HD - Chorea. Hence, normalising the neurotrophin receptor signalings can be and will be an effective treatment option due to the potential ameliorative effects that it may have on the neuropathological, and physiological conditions of the HD patients.

Introduction

Huntington's Disease (HD) is an inherited neurodegenerative disorder (Roos, 2010). It is an autosomal dominant disease caused by an expansion of CAG repeats (codon that codes for the amino acid glutamine) within the first exon of the Huntingtin (HTT) gene in chromosome 4.

Normal HTT gene encodes for the normal HTT protein, while the expanded HTT gene codes for a mutant HTT protein (mHTT). The expression of HTT protein is ubiquitous, and that it is not restricted to the brain (Marques Sousa & Humbert, 2013). However, its physiological function is still unclear.

HTT protein contains three domains, which includes the N-terminal, polyglutamine tract (polyQ tract), and a carboxyl-terminal. HD patients usually have over 35 CAG repeats within the first exon of the HTT gene, resulting in an expanded polyQ tract. Interestingly, elderly patients with 36 to 40 CAG repeats can be asymptomatic, which demonstrates how mutations may not always be fully penetrant (Rubinsztein et al., 1996).

The expanded polyQ may cause mHTT to fold abnormally, forming misfolded mHTTs that are relatively toxic (Shacham et al., 2019). In this case, mHTT may be spliced by endonucleases to form N-terminal fragments (Tabrizi et al., 2020). Aggregation of these mHTT fragments may lead to transcriptional deregulation or proteasome impairment, affecting neuronal survival (Arrasate & Finkbeiner, 2012). However, mHTT protein aggregates are also known to possess several benefits including the removal of toxic and misfolded proteins (Mcfarland & Cha, 2011). The precise role of protein aggregates remains unclear.

This disease largely affects a particular population of neurons found within the basal ganglia, a major structure in the brain which is responsible for motor control and other cognitive actions. One of the major components of the basal ganglia is the striatum, which is made up mainly of GABAergic neurons called medium spiny neurons (MSNs). There are two types of MSNs, identified by the kind of dopamine receptor they express, either D1R or D2R. These two kinds of neurons have different functions. D1R MSNs, also known as MSNs of the direct pathway, connect by way of many other structures to the motor cortex, where it induces execution of motor programs. D2Rs MSNs, also known as MSNs of the indirect pathway, also connect to the motor cortex where they tonically inhibit activation of motor programs. These two pathways work in concert to only activate

the most adaptive motor programs for a given situation (Knierim, 2020). However, in HD patients, the neurons of the indirect pathway will undergo apoptosis, leading in a loss of inhibition in the motor cortex and a consequent overactivation of non-adaptive motor programs. Therefore, HD patients typically suffer from chorea, uncontrolled involuntary movements.

Apart from chorea, other clinical manifestation of HD includes motor impairment, and cognitive impairment (Ross et al., 2014). Therefore, HD can be considered a debilitating disease.

Mechanisms for Striatal Atrophy

Chorea is the most significant clinical symptom of HD patients. As mentioned, it is caused by striatal atrophy. In this section, the molecular mechanisms underlying striatal atrophy will be explored.

Neurotrophins

Neurotrophins is a family of proteins that are responsible for neuronal growth, survival, and development in the central nervous system (CNS) and the peripheral nervous system (PNS) (Sahay et al., 2017). Important neurotrophins include nerve growth factors (NGF), brain-derived neurotrophic factors (BDNF), neurotrophin-3 (NT3), and neurotrophin-4 (NT4). In fact, NGF was the first neurotrophin to be discovered. The discovery and isolation of NGF by Nobel Laureate Rita Levi-Montalcini and Stanley Cohen was revolutionary (Garfield, 1987). It has set the stage for further discovery of other neurotrophins and their roles in various neurodegenerative diseases including Alzheimer's disease (AD) and HD (Jiao et al., 2016; Zuccato & Cattaneo, 2007). Different neurotrophins bind to different receptors and stimulate or inhibit different types of actions. This research paper will focus on the roles of BDNF and its receptors in the development and progression of HD.

Brain-Derived Neurotrophic Factor (BDNF)

BDNF is a neurotrophin that is encoded by the BDNF gene. It was first isolated from pig brains by Yves-Alain Barde and Hans Thoenen in 1989 (Kowiański et al., 2018). It can be found in the CNS and other locations such as saliva and the collecting duct of the kidney (Mandel et al., 2011; Tao et al., 2018). However, BDNF expression is the most abundant in the CNS as BDNF messenger RNA (mRNA) and thus BDNF protein are more highly expressed in the neocortex, hippocampus, amygdala, and cerebellum than other peripheral tissues (Benarroch, 2015). In the brain, BDNF is required for many different functions. For instance, BDNF produced by cortical neurons will be transported anterogradely to the MSNs of the basal ganglia, promoting the development and maintenance of the MSNs of the indirect pathways (Baydyuk & Xu, 2014). Since BDNF is a protein, it is too big to be transported across the plasma membrane. Therefore, it must be bound to transmembrane receptors. The two classes of glycosylated receptors that BDNF binds to are tyrosine receptor kinase B (TrkB) and p75 neurotrophin receptor (p75NTR) (Gong et al., 2008).

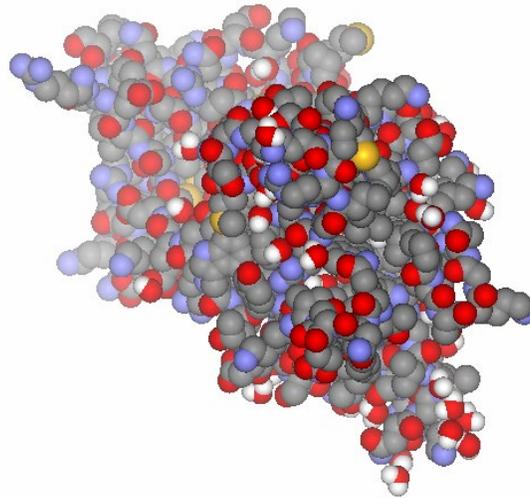


Figure 1. A figure of BDNF (Microswitch, 2007).

Tyrosine Receptor Kinase B (TrkB)

TrkB is a transmembrane protein localised on the plasma membrane on the axon end. Not only is it a receptor for BDNF, but also a receptor for NT4. TrkB, like other Trk receptors, are type I receptor tyrosine kinases (RTKs). It has an extracellular ligand-binding domain and an intracellular kinase domain (Ivanisevic & Saragovi, 2013; Li & Hristova, 2006).

It is believed that TrkB exists in a monomer-dimer equilibrium, and that ligand binding to the extracellular ligand-binding domain stabilises the formation of the active dimers that conduct biochemical signals in the cell. When neurotrophins such as BDNF and NT4 bind to extracellular ligand-binding site of the TrkB, the TrkB monomer will recruit another inactive TrkB to undergo dimerisation. This lateral dimerisation brings two TrkB monomers in close contact to form non-covalently associated dimer. The interaction between the two cytoplasmic domains results in intermolecular autophosphorylation of the receptors, where phosphate groups are added onto the cytoplasmic tails of each monomer. This activates the receptors as it allows the formation of binding sites for adaptor proteins such as RAS, Shc, growth factor, triggering signaling cascades (Li & Hristova, 2006; Schlessinger, 2000; Zhang et al., 2006).

Mature TrkB activates three downstream phosphorylation cascades. One includes the recruitment of RAS to TrkB, which activates mitogen-activated protein kinase (MAPK) and hence the extracellular signal-regulated kinases (ERK). The second cascade is triggered by recruitment of adaptor proteins Shc and growth factor receptor-bound protein 2 (Grb2). Shc and Grb2 activate phosphatidylinositol 3-kinase (PI3K), and subsequently protein kinase B (AKT). The third signaling cascade is related to the enzymatic activities of phospholipase C (PLC). PLC interacts with the phospholipid component on the cell membrane known as phosphatidylinositol 4,5-bisphosphate (PIP₂). PLC cleaves and hydrolyses PIP₂ into two well-known second messengers – diacylglycerol (DAG) and inositol triphosphate (IP₃) (Harras et al., 2020). DAG activates protein kinase C (PKC), whereas IP₃ binds onto the transmembrane receptors on the smooth endoplasmic reticulum, allowing a release of calcium ions into the cytoplasm. These cascades - MAPK/ERK, PI3K/AKT, and PLC result in both spine integrity and cell growth and survival, thus BDNF is said to have neuroprotective and growth-promoting effects (Simmons, 2017).

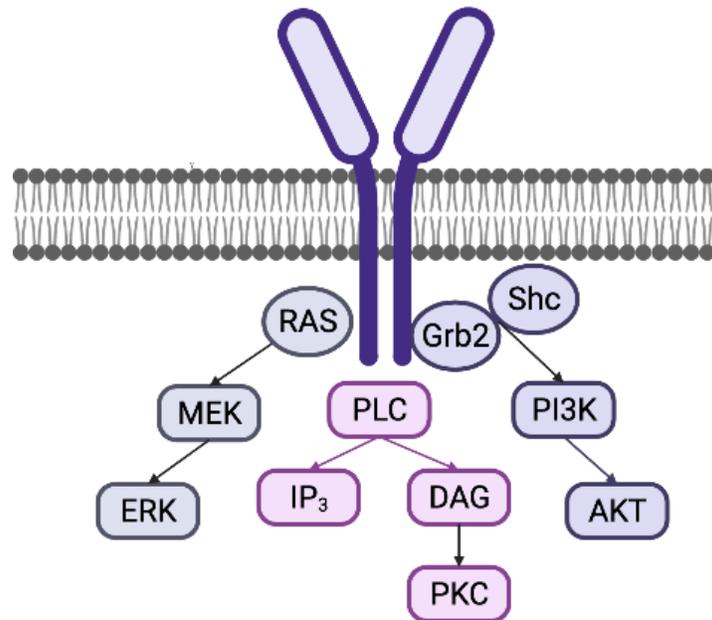


Figure 2. A figure showing the signaling cascades of TrkB receptor, created in BioRender. p75 Neurotrophin Receptor (p75NTR)

BDNF and NT4 also bind to p75NTR. p75NTR belongs to the family of tumour necrosis factor receptors. It has an extracellular ligand-binding domain and an intracellular death domain (Ivanisevic & Saragovi, 2013). Unlike TrkB, its cytoplasmic domain has no catalytic activity. Apart from BDNF and NT4, all other neurotrophins and proneurotrophins can also bind onto p75NTR, activating apoptosis pathways or stimulating cell proliferation (Gong et al., 2008).

The apoptosis pathway is triggered by the recruitment of proteins including neurotrophin receptor interacting factor (NRIF), neurotrophin receptor interacting MAGE homolog (NRAGE), and tumour necrosis factor associated factors (TRAFs). These proteins trigger the activation of the c-Jun N-terminal kinase (JNK) signaling, leading to cell death (Simmons, 2017).

Interesting, p75NTR can also act as co-receptor for Trk receptors. In this case, p75NTR will stimulate some completely different, virtually contradictory activities. When it physically interacts with Trk receptors, it performs three main functions – altering the ligand-binding specificity of Trk receptors, enhancing the binding affinity of Trk receptors, and enhancing the ligand-induced tyrosine kinase activity of Trk receptors (Greene & Kaplan, 1995). Therefore, Trk-p75NTR interactions can also enhance the activation of the three neuroprotective and growth-promoting cascades.

Furthermore, p75NTR also interacts with sortilin. This p75NTR-sortilin complex induces apoptosis. This mechanism is usually upregulated following a neuronal injury (Jansen et al., 2007).

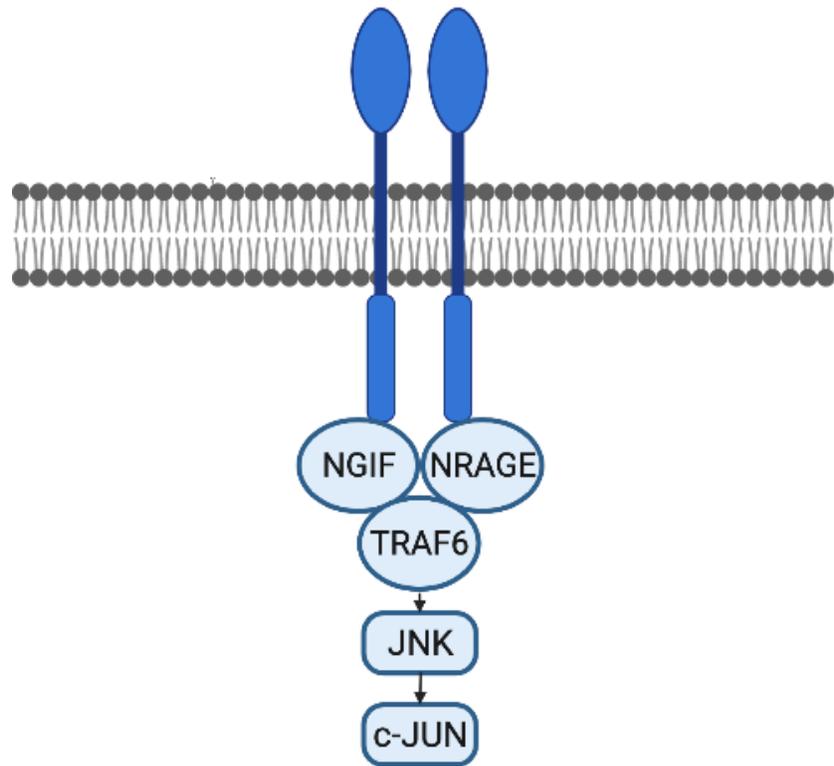


Figure 3. A figure showing the signaling cascades of p75NTR, created in BioRender

Signalings in Huntington’s Disease Patients

There are two hypotheses regarding the roles of neurotrophin signaling pathways in the development of striatal atrophy in patients with Huntington’s disease. They are BDNF deficiency, and imbalance of p75NTR/TrkB protein expression.

BDNF Deficiency

Since BDNF plays a significant role in the regulation of cell survival, deficiency of BDNF can be detrimental to cellular health. For example, since BDNF produced in cortical axons is required for regulating striatal neuron size, deficiency of BDNF implies a reduction in the binding of BDNF to the TrkB receptors on the striatal MSNs, resulting in reduced activation of neuronal survival pathways, possibly leading to the deaths of striatal MSNs. Since this is particularly evident in patients with Huntington’s disease, different studies are done to investigate the relationship behind the deficiency of BDNF in patients and mHTT.

Firstly, a study has compared the axonal movement of BDNF in cortical neurons in wild type mice (WT) and mice expressing full-length human mHTT (BACHD). Kymograph results have shown that there were significantly more pausing in BACHD neurons than in WT neurons. Therefore, it can be concluded that mHTT has a potential role in disrupting axonal transport of BDNF, and that it may impact the delivery of BDNF to the striatum (Zhao et al., 2016).

Moreover, another study has also shown a 24% reduction of BDNF exon II mRNA level in 8-week-old HD mice, and a 58.7% reduction of BDNF exon II mRNA level in 12-week-old HD mice (Zuccato et al., n.d.). A progressive reduction of mRNA demonstrates a progressive decrease in transcription of BDNF gene, implying a reduction of BDNF gene expression. Therefore, a negative correlation between mHTT and BDNF expression is established.

In conclusion, it is hypothesised that mHTT exerts an effect on both the expression of BDNF and the transport of BDNF to its target, subsequently reducing the activation of TrkB receptors and the downstream signaling cascades responsible for neuroprotective and neuronal growth. Since BDNF is particularly crucial for regulating the survival of MSNs, deficiency may then trigger deaths of MSNs, progressively resulting in striatal atrophy.

Imbalance of p75NTR/TrkB Protein Expression

Whether BDNF can perform its neuroprotective and neuronal growth functions depends largely on the actions and thus the signaling induced by the BDNF receptors. Therefore, apart from BDNF, the expression of its receptors also plays a role in regulating neuronal survival. For instance, when there is a decrease in the TrkB receptor, there will be less ligand-binding sites available for BDNF to bind to, and hence there will be less activations of neuronal survival pathways, possibly leading to neuronal death. Conversely, an increase in the expression of p75NTR indicates an increase in activation of apoptosis pathways, further promoting apoptosis. Therefore, researchers have also investigated the relationships between mHTT, expressions of TrkB and p75NTR, and the survival/death of MSNs in the striatum.

A study on TrkB expressions has been performed on HD mouse models (Ginés et al., 2006). This study concludes that, there was a reduction in TrkB expression in the striatum and the cortex of HD mice, and that the overall reduction was caused by the reduction in TrkB mRNA, instead of cell deaths or the aggregation and cleavage by mHTT. Therefore, this suggests that, the presence of mHTT may lead to a reduction in the expression of TrkB gene, leading to a reduction in TrkB in the striatum and the cortex.

The reduction of TrkB expression is further complicated by the increase in p75NTR expression. Research has found an increase in p75NTR in the caudate (Zuccato et al., 2008). This imbalance of p75NTR/TrkB protein expressions will ultimately lead to a decrease in phosphorylation of ERK and AKT, and an increase in JNK phosphorylation, reducing pro-survival signaling while increasing pro-apoptosis signaling (Brito et al., 2013).

In addition, cells with mHTT are also found to have reduced the levels of Shc adapter proteins binding to active TrkB (Ginés et al., 2010). In this case, the PI3K/AKT pathway will be affected, affecting the signaling for neuronal survival, potentially leading to neuronal death.

In conclusion, mHTT is found to decrease TrkB gene expression and signaling and increase p75NTR expression. This subsequently downregulates the signaling pathways for neuronal survival while upregulating the signaling pathways for apoptosis. Therefore, this provides explanation to why striatal neurons, such as MSNs, are prone to degeneration under the influence of mHTT.

Treatment Strategies Targeting Neurotrophic Signaling Pathways

When considering the treatment strategies for Huntington's disease, a lot of attention has been focused on removing mHTT. For example, Tominersen, an antisense oligonucleotide in the clinical trial phase III, is developed to target and remove the mHTT. This is a possible treatment option, but it is worth noting that it does not help with restoring the neurotrophin imbalance in HD patients. Novel strategies targeting BDNF receptor signaling pathways not only are innovative but also target the molecular causes of the striatal atrophy, including BDNF deficiency and the imbalance of p75NTR/TrkB protein expression.

BDNF-Based Strategies

Regarding the deficiency of BDNF in the striatum, one of the most direct approaches would be to deliver exogenous BDNF. Since BDNF is a peptide, oral administration is not an option, as it will be easily digested by peptidases in the gastrointestinal tract, lowering the bioavailability. Moreover, it is also important to note that BDNF can only stay in the blood for as long as 60 minutes (Pan et al., 1998), and that BDNF is too big to penetrate the blood-brain barrier. Therefore, if BDNF is administered through systematic delivery such as intravenous approaches, the instability of BDNF in the blood and the low blood-brain barrier permeability will adversely reduce the bioavailability, limiting its effects. In this case, more invasive approaches of administering BDNF such as intrathecal injection is required. With intrathecal injection, BDNF can be injected into the cerebrospinal fluid via lumbar puncture, which then allows BDNF to travel along the cerebrospinal fluid to reach striatum.

Intrathecal injection of BDNF for HD is not widely studied. However, this is widely recognized and studied for amyotrophic lateral sclerosis (ALS). Pre-clinical trials for intrathecal infusion of BDNF for ALS show that when BDNF is administered into the subarachnoid space, a specific supply of BDNF into spinal motor neurons can result (Dittrich et al., 1996). Furthermore, in a randomized, double-blind study on the efficacy of intrathecal administration of recombinant methionyl human BDNF into subarachnoid space for ALS patients, results confirm that doses less than 150 microg/day is feasible. However, adverse effects such as changes in mood, behaviours, and sleep disturbance will be resulted when doses are at a level greater than 150 microg/day (Ochs et al., 2000). However, the presence of these adverse effects is explainable. Research has concluded that, due to low specificity and inability to control the doses in intrathecal injections, adverse side effects such as infections, headaches, damage to nerves and the spinal cord can be easily resulted (Staats, 2008). Apart from adverse side-effects, BDNF is also shown to have a short half-life of 62.7 minutes in the cerebrospinal fluid, and thus cannot be deemed to be significantly long enough to be used as a drug therapeutic option. Interestingly, when BDNF is covalently attached to polyethylene glycol, the half-life can be increased to 167 minutes (Soderquist et al., 2009). However, a half-life of 167 minutes (equivalent to around 2.8 hours) can still be considered short. Thus, if this approach is adopted, there will be a need for a high frequency of administration to maintain the level of BDNF in the cerebrospinal fluid. Yet, according to research on administering exogenous BDNF for rats with spinal cord injury, persistent exposure to BDNF could lead to maladaptive adverse side effects, such as enhanced spasticity and sensitivity to heat (Boyce et al., 2012). Consequently, in order to make administration of BDNF a feasible option, more investigations must be done to minimise the side effects while maximising its efficacy.

TrkB-Based Strategies

There have been multiple investigations dedicated to strategically increase the activation of TrkB receptors, in the hope of increasing the activation of the three main downstream pathways - MAPK/ERK, PI3K/AKT, and PLC, stimulating neuronal proliferation. These strategies include the use of TrkB agonists and the use of monoclonal antibodies.

TrkB Agonists

The use of TrkB agonists to activate TrkB dimerisation, autophosphorylation and the subsequent downstream signaling pathways is one of the ways proposed. TrkB agonists include 7,8-Dihydroxyflavone (7,8-DHF), amitriptyline, imipramine, fluoxetine, LM22A-4.

7,8-DHF can activate TrkB as well as BDNF, and that its apoptosis inhibitory activities may even exceed that of BDNF (Jang, Liu, Yepes, et al., 2010). Preclinical experiments of 7,8-DHF on R6/1 mice has

confirmed its efficacy in restoring the TrkB/p75 imbalance and preventing cell death through activating the PLC pathway, resulting in functional improvements (Jang, Liu, Yepes, et al., 2010). In terms of toxicity, 7,8-DHF treatment did not trigger any apparent toxicity in mice during preclinical studies (C. Liu et al., 2016). However, in terms of its chemical structure, it has a reactive catechol group that is prone to reactions such as oxidation, glucuronidation, sulfation, and methylation. Therefore, 7,8-DHF has low bioavailability and suboptimal brain exposure (X. Liu et al., 2013). Due to this limitation, scientists have further developed this TrkB agonist. As the catechol group is the pharmacophore, a prodrug strategy was taken to reduce the reactivity of the drug in the plasma, improving its bioavailability. This eventually led to the development of a compound R13, a prodrug of 7,8-DHF (Chen et al., 2018). According to the research paper, it can be hydrolysed into an intermediate (T1), then 7,8-DHF in liver microsomes or plasma. Furthermore, the paper also confirms that R13 has a very long half-life, and that it improves 7,8-DHF's oral bioavailability from 4.6% to around 10.5%. Therefore, this compound can be deemed to be a potential candidate for restoring the imbalance of signaling in HD patients.

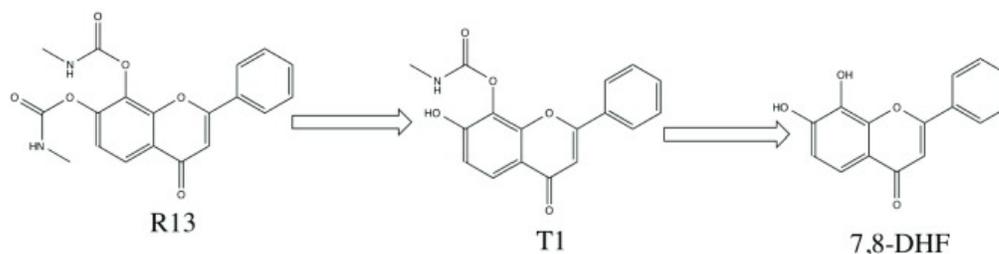


Figure 4. A figure showing the chemical skeletal formula of R13, T1, and 7,8-DHF (Chen et al., 2018).

Antidepressant drug Amitriptyline can activate TrkB dimerisation, autophosphorylation and the downstream signalings including MAPK/ERK, PI3K/AKT, and PLC in WT mouse brain. However, other antidepressant drugs such as imipramine and fluoxetine are found have a better effect than amitriptyline in triggering TrkB activation (Jang et al., 2009). Although studies have shown that antidepressant drugs can activate TrkB receptors (Jang et al., 2009; Rantamäki et al., 2011), extensive studies on their role in stimulating cell survival in HD mice have not been done. Therefore, their efficacy as a HD treatment strategy remains unknown.

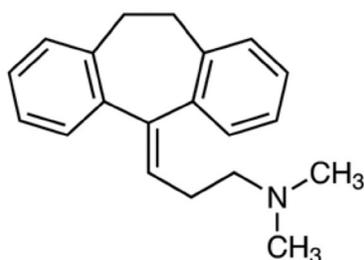


Figure 5. A figure showing the chemical skeletal formula of Amitriptyline (*Amitriptyline Hydrochloride*, n.d.)

LM22A-4 is a partial TrkB agonist. In vitro studies have shown that LM22A-4 is able to trigger an increase in activation of TrkB, MAPK/ERK, PI3K/AKT pathways in hippocampus and striatum, matching BDNF's efficacy in preventing neuronal death (Massa et al., 2010). Administration of LM22A-4 to R6/2 and BACHD mice was also performed (Simmons et al., 2013). According to the results, LM22A-4 was associated with an increase in TrkB activation and two of the three major signaling pathways – PI3K/AKT, and PLCg

pathways. Reduction in motor impairment and other HD-associated mechanisms such as mHTT protein aggregates were also recorded. This suggests the therapeutic potential of LM22A-4 in regulating the neurotrophic signalings in HD patients.

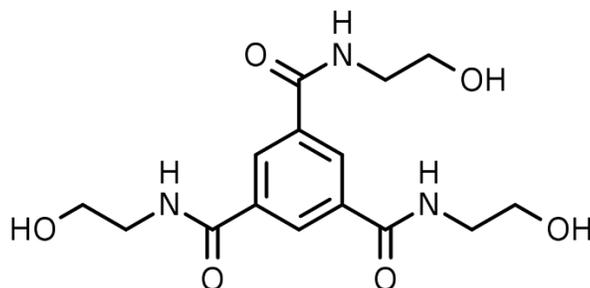


Figure 6. A figure showing the chemical skeletal formula of LM22A-4 (Aethyta, 2016)

Endogenous N-acetylserotonin, apart from just being a precursor for melatonin, it has been found to be a TrkB agonist (Zhou et al., 2014). According to an in vitro study, N-acetylserotonin stimulated the activation of TrkB and the MAPK/ERK, PI3K/AKT pathways in the retina and hippocampus of wild-type C3H/f^{+/+} mice (Jang, Liu, Pradoldej, et al., 2010). However, due to the lack of research, it is still unclear whether exogenous N-acetylserotonin will be able to reach the striatum to exert ameliorative effects on BACHD mice.

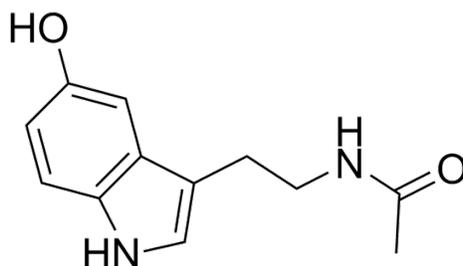


Figure 7. A figure showing the chemical skeletal formula of N-acetylserotonin (Chemgir1131, 2010)

Monoclonal Antibodies (mAb)

Apart from the chemically synthesised molecules, the use of monoclonal antibodies (mAb) is also proposed. mAb has been shown to be capable of inducing dimerisation, autophosphorylation and subsequent downstream signaling pathways of trkB. For instance, upon investigating on mice models, researchers found that mAb 1D7 can induce the activation of TrkB and prevent retinal ganglion cells from death in retinal disease (Bai et al., 2010).

A recent study using cortico-striatal co-cultures has also confirmed the effectiveness of 29D7 and 38B8 mAbs from Pfizer in activating TrkB and downstream pathways. The introduction of these monoclonal mAb is also confirmed to be able to reduce cell loss, reaching a neuroprotective effect in the co-culture with mHTT expressions (Todd et al., 2014). However, these monoclonal antibodies have not been tested in HD mice models. Thus, the efficacy remains unknown. Furthermore, due to their peptide nature, its ability to penetrate the blood-brain barrier is questionable as well. Would intrathecal injections be effective enough to allow them to be delivered to the striatum? This remains the biggest challenge for the monoclonal antibodies.

p75NTR-Based Strategies

To restore the imbalance of TrkB and p75 signaling, solutions to downregulate p75NTR are needed as well.

An in vitro study has been found that LM11A compounds only act through p75NTR, and that they do not act through Trk receptors. Although p75NTR is known to induce cell death, the binding of LM11A-24 and -31 onto p75NTR is seen to be able to inhibit its ability to induce cell death (Massa et al., 2006), as it is believed that these LM11A compounds inhibit the binding of the precursor of NGF (pro-NGF) to the extracellular domain of p75, and hence inhibiting the proNGF-induced apoptosis of oligodendrocytes in cultures (Longo & Massa, 2013).

LM11A-31 is able to restore striatal PI3K/AKT and MAPK/ERK while inhibiting JNK, hence reducing inflammation, increasing spine integrity, and improving motor and cognitive functions in R6/2 and BACHD mice (Simmons et al., 2013). Since LM11A-31 is a small, non-polar, non-peptide molecule, it exhibits significant oral uptake and blood-brain penetration (Knowles et al., 2013; Massa et al., 2006). Thus, it has a strong therapeutic potential for HD patients. In fact, LM11A-31 is currently in the Phase IIa clinical trial for AD (*Study of LM11A-31-BHS in Mild-Moderate AD Patients*, 2017). Currently, there are scientists developing effective pharmacodynamic biomarkers to detect the therapeutic effects of LM11A-31 (Simmons et al., 2021). This demonstrates how scientists are working towards putting the drug in HD clinical trials.

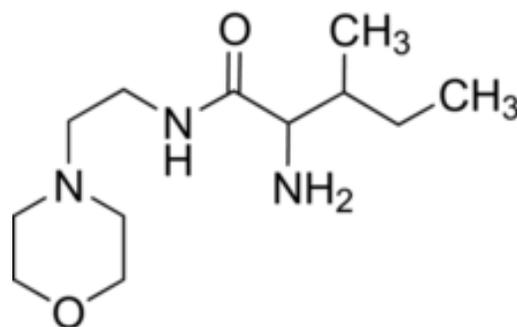


Figure 8. A figure showing the chemical skeletal formula of LM11A-31 (*LM11A-31 Dihydrochloride*, n.d.)

Conclusion

In conclusion, BDNF deficiency and the imbalance of the p75NTR/TrkB expression are responsible for striatal atrophy in HD patients, which collectively provide a molecular explanation to the most significant hallmark symptom of HD - Chorea.

In this paper, BDNF-based, TrkB-based, and p75NTR-based approaches to restore healthy neurotrophin signaling are explored. It is worth noting that most of the proposed treatments are not fully developed. Scientists are still modifying the drugs to maximise their efficacies, while minimising the limitations such as the presence of adverse side-effects, short half-life, low bioavailability due to high solubility in plasma or low permeability across the blood-brain barrier.

With enough pre-clinical and clinical research, normalising the neurotrophin receptor signalings can be and will be an effective treatment option due to the potential ameliorative effects that it may have on the neuropathological, and physiological conditions of the HD patients.

Moreover, based on above discussions, it is the opinion of this author that TrkB-based and p75NTR-based strategies have equal potential, while BDNF-based strategies are less ideal in terms of feasibility and efficacy. Moving forward, more attention should be given to strategies that restore the imbalance of p75NTR/TrkB protein expression, as it is ultimately TrkB and p75NTR that play the most crucial role in activating MAPK/ERK, PI3K/AKT, PLC pathways and inhibiting the JNK signaling in the promotion of neuronal growth and survival.

Yet, normalising neurotrophin signaling is just one of the developing therapeutic solutions for HD patients. Its high efficacy should not be a factor that jeopardises the development of other therapeutic options, rather, it should be encouraging for field of HD therapeutics. When they become available, they should be used in conjunction with other therapeutic options in order to provide the best possible holistic care for HD patients.

Acknowledgment

I would like to thank my supervisor Mr. Kenneth Kuhn deeply for introducing me to the marvels of the neuroscience and guiding me throughout the process of writing up this research paper.

I also thank Horizon Academic for providing this valuable opportunity for me to practice college-level writings.

Last but not the least, I thank those who have given me endless support in pursuing my dreams – my parents.

Bibliography

- Aethyta. (2016, January 9). *LM22A-4 structure*. https://commons.wikimedia.org/wiki/File:LM22A-4_structure.svg
- Amitriptyline Hydrochloride*. (n.d.). TCI . Retrieved November 28, 2021, from <https://www.tcichemicals.com/HK/en/p/A0908>
- Arrasate, M., & Finkbeiner, S. (2012). Protein aggregates in Huntington's disease. In *Experimental Neurology* (Vol. 238, Issue 1, pp. 1–11). <https://doi.org/10.1016/j.expneurol.2011.12.013>
- Bai, Y., Xu, J., Brahimi, F., Zhuo, Y., Sarunic, M. v., & Uri Saragovi, H. (2010). An agonistic TrkB mAb causes sustained TrkB activation, delays RGC death, and protects the retinal structure in optic nerve axotomy and in glaucoma. *Investigative Ophthalmology and Visual Science*, *51*(9), 4722–4731. <https://doi.org/10.1167/iovs.09-5032>
- Baydyuk, M., & Xu, B. (2014). BDNF signaling and survival of striatal neurons. In *Frontiers in Cellular Neuroscience* (Vol. 8, Issue AUG). <https://doi.org/10.3389/fncel.2014.00254>
- Benarroch, E. E. (2015). *CLINICAL IMPLICATIONS OF NEUROSCIENCE RESEARCH Section Editor Brain-derived neurotrophic factor Regulation, effects, and potential clinical relevance*.
- Boyce, V. S., Park, J., Gage, F. H., & Mendell, L. M. (2012). Differential effects of brain-derived neurotrophic factor and neurotrophin-3 on hindlimb function in paraplegic rats. *European Journal of Neuroscience*, *35*(2), 221–232. <https://doi.org/10.1111/j.1460-9568.2011.07950.x>
- Brito, V., Puigdellívol, M., Giralt, A., del Toro, D., Alberch, J., & Ginés, S. (2013). Imbalance of p75NTR/TrkB protein expression in Huntington's disease: Implication for neuroprotective therapies. *Cell Death and Disease*, *4*(4). <https://doi.org/10.1038/cddis.2013.116>
- Chemgirl131. (2010, March 29). *N-Acetylserotonin*. <https://commons.wikimedia.org/wiki/File:N-Acetylserotonin.png>
- Chen, C., Wang, Z., Zhang, Z., Liu, X., Kang, S. S., Zhang, Y., & Ye, K. (2018). The prodrug of 7,8-dihydroxyflavone development and therapeutic efficacy for treating Alzheimer's disease. *Proceedings of the National Academy of Sciences of the United States of America*, *115*(3), 578–583. <https://doi.org/10.1073/pnas.1718683115>
- Dittrich, F., Gu, G., Ochs, G., Große-Wilde, A., Berweiler, U., Yan, Q., Miller, J. A., Toyka, K. v., & Sendtner, M. (1996). *Pharmacokinetics of Intrathecally Applied BDNF and Effects on Spinal Motoneurons*.

- Garfield, E. (1987). *Stanley Cohen's and Rita Levi-kfontdcini's Discoveries of Growth Factors Lead to 1986 Nobel in Medicine*. 10, 106.
- Ginés, S., Bosch, M., Marco, S., Gavaldà, N., Díaz-Hernández, M., Lucas, J. J., Canals, J. M., & Alberch, J. (2006). Reduced expression of the TrkB receptor in Huntington's disease mouse models and in human brain. *European Journal of Neuroscience*, 23(3), 649–658. <https://doi.org/10.1111/j.1460-9568.2006.04590.x>
- Ginés, S., Paoletti, P., & Alberch, J. (2010). Impaired TrkB-mediated ERK1/2 activation in huntington disease knock-in striatal cells involves reduced p52/p46 Shc expression. *Journal of Biological Chemistry*, 285(28), 21537–21548. <https://doi.org/10.1074/jbc.M109.084202>
- Gong, Y., Cao, P., Yu, H. J., & Jiang, T. (2008). Crystal structure of the neurotrophin-3 and p75NTR symmetrical complex. *Nature*, 454(7205), 789–793. <https://doi.org/10.1038/nature07089>
- Greene, L. A., & Kaplan, D. R. (1995). Early events in neurotrophin signalling via Trk and p75 receptors. In *Current Opinion in Neurobiology* (Vol. 5).
- Harraz, O. F., Hill-Eubanks, D., & Nelson, M. T. (2020). PIP2: A critical regulator of vascular ion channels hiding in plain sight. *Proceedings of the National Academy of Sciences of the United States of America*, 117(34), 20378–20389. <https://doi.org/10.1073/pnas.2006737117>
- Ivanisevic, L., & Saragovi, H. U. (2013). Neurotrophins. In *Handbook of Biologically Active Peptides* (pp. 1639–1646). Elsevier Inc. <https://doi.org/10.1016/B978-0-12-385095-9.00224-4>
- Jang, S. W., Liu, X., Chan, C. B., Weinschenker, D., Hall, R. A., Xiao, G., & Ye, K. (2009). Amitriptyline is a TrkA and TrkB Receptor Agonist that Promotes TrkA/TrkB Heterodimerization and Has Potent Neurotrophic Activity. *Chemistry and Biology*, 16(6), 644–656. <https://doi.org/10.1016/j.chembiol.2009.05.010>
- Jang, S. W., Liu, X., Pradoldej, S., Tosini, G., Chang, Q., Iuvone, P. M., & Ye, K. (2010). N-acetylserotonin activates TrkB receptor in a circadian rhythm. *Proceedings of the National Academy of Sciences of the United States of America*, 107(8), 3876–3881. <https://doi.org/10.1073/pnas.0912531107>
- Jang, S. W., Liu, X., Yepes, M., Shepherd, K. R., Miller, G. W., Liu, Y., Wilson, W. D., Xiao, G., Bianchi, B., Sun, Y. E., & Ye, K. (2010). A selective TrkB agonist with potent neurotrophic activities by 7,8-dihydroxyflavone. *Proceedings of the National Academy of Sciences of the United States of America*, 107(6), 2687–2692. <https://doi.org/10.1073/pnas.0913572107>
- Jansen, P., Giehl, K., Nyengaard, J. R., Teng, K., Liubinski, O., Sjoegaard, S. S., Breiderhoff, T., Gotthardt, M., Lin, F., Eilers, A., Petersen, C. M., Lewin, G. R., Hempstead, B. L., Willnow, T. E., & Nykjaer, A. (2007). Roles for the pro-neurotrophin receptor sortilin in neuronal development, aging and brain injury. *Nature Neuroscience*, 10(11), 1449–1457. <https://doi.org/10.1038/nn2000>
- Jiao, S. S., Shen, L. L., Zhu, C., Bu, X. L., Liu, Y. H., Liu, C. H., Yao, X. Q., Zhang, L. L., Zhou, H. D., Walker, D. G., Tan, J., Götz, J., Zhou, X. F., & Wang, Y. J. (2016). Brain-derived neurotrophic factor protects against tau-related neurodegeneration of Alzheimer's disease. *Translational Psychiatry*, 6(10). <https://doi.org/10.1038/tp.2016.186>
- Knierim, J. (2020, October 20). *Basal Ganglia*. Neuroscience Online. <https://nba.uth.tmc.edu/neuroscience/m/s3/chapter04.html>
- Knowles, J. K., Simmons, D. A., Nguyen, T. V. v., vander Griend, L., Xie, Y., Zhang, H., Yang, T., Pollak, J., Chang, T., Arancio, O., Buckwalter, M. S., Wyss-Coray, T., Massa, S. M., & Longo, F. M. (2013). A small molecule p75NTR ligand prevents cognitive deficits and neurite degeneration in an Alzheimer's mouse model. *Neurobiology of Aging*, 34(8), 2052–2063. <https://doi.org/10.1016/j.neurobiolaging.2013.02.015>
- Kowiański, P., Lietzau, G., Czuba, E., Waśkow, M., Steliga, A., & Moryś, J. (2018). BDNF: A Key Factor with Multipotent Impact on Brain Signaling and Synaptic Plasticity. In *Cellular and Molecular*

- Neurobiology* (Vol. 38, Issue 3, pp. 579–593). Springer New York LLC.
<https://doi.org/10.1007/s10571-017-0510-4>
- Li, E., & Hristova, K. (2006). Role of receptor tyrosine kinase transmembrane domains in cell signaling and human pathologies. In *Biochemistry* (Vol. 45, Issue 20, pp. 6241–6251).
<https://doi.org/10.1021/bi060609y>
- Liu, C., Chan, C. B., & Ye, K. (2016). 7,8-dihydroxyflavone, a small molecular TrkB agonist, is useful for treating various BDNF-implicated human disorders. In *Translational Neurodegeneration* (Vol. 5, Issue 1). BioMed Central Ltd. <https://doi.org/10.1186/s40035-015-0048-7>
- Liu, X., Qi, Q., Xiao, G., Li, J., Luo, H. R., & Ye, K. (2013). O-methylated metabolite of 7,8-dihydroxyflavone activates TrkB receptor and displays antidepressant activity. *Pharmacology*, *91*(3–4), 185–200. <https://doi.org/10.1159/000346920>
- LM11A-31 dihydrochloride*. (n.d.). Sigma-Aldrich. Retrieved November 28, 2021, from <https://www.sigmaaldrich.com/HK/zh/product/sigma/sml0664>
- Longo, F. M., & Massa, S. M. (2013). Small-molecule modulation of neurotrophin receptors: A strategy for the treatment of neurological disease. In *Nature Reviews Drug Discovery* (Vol. 12, Issue 7, pp. 507–525). <https://doi.org/10.1038/nrd4024>
- Mandel, A. L., Ozdener, H., & Utermohlen, V. (2011). Brain-derived neurotrophic factor in human saliva: Elisa optimization and biological correlates. *Journal of Immunoassay and Immunochemistry*, *32*(1), 18–30. <https://doi.org/10.1080/15321819.2011.538625>
- Marques Sousa, C., & Humbert, S. (2013). Huntingtin: Here, there, everywhere! *Journal of Huntington's Disease*, *2*(4), 395–403. <https://doi.org/10.3233/JHD-130082>
- Massa, S. M., Xie, Y., Yang, T., Harrington, A. W., Mi, L. K., Sung, O. Y., Kraemer, R., Moore, L. A., Hempstead, B. L., & Longo, F. M. (2006). Small, nonpeptide p75NTR ligands induce survival signaling and inhibit proNGF-induced death. *Journal of Neuroscience*, *26*(20), 5288–5300. <https://doi.org/10.1523/JNEUROSCI.3547-05.2006>
- Massa, S. M., Yang, T., Xie, Y., Shi, J., Bilgen, M., Joyce, J. N., Nehama, D., Rajadas, J., & Longo, F. M. (2010). Small molecule BDNF mimetics activate TrkB signaling and prevent neuronal degeneration in rodents. *Journal of Clinical Investigation*, *120*(5), 1774–1785. <https://doi.org/10.1172/JCI41356>
- Mcfarland, K. N., & Cha, J. H. J. (2011). Molecular biology of Huntington's disease. In *Handbook of Clinical Neurology* (Vol. 100). <https://doi.org/10.1016/B978-0-444-52014-2.00003-3>
- Microswitch. (2007, January 1). *Brain-derived neurotrophic factor*. https://commons.wikimedia.org/wiki/File:Brain-derived_neurotrophic_factor_-_PDB_id_1BND.png
- Ochs, G., Penn, R. D., York, M., Giess, R., Beck, M., Tonn, J., Haigh, J., Malta, E., Traub, M., Sendtner, M., & Toyka, K. v. (2000). A phase I/II trial of recombinant methionyl human brain derived neurotrophic factor administered by intrathecal infusion to patients with amyotrophic lateral sclerosis. *Amyotrophic Lateral Sclerosis*, *1*(3), 201–206. <https://doi.org/10.1080/14660820050515197>
- Pan, W., Banks, W. A., Fasold, M. B., Bluth, J., & Kastin, A. J. (1998). Transport of brain-derived neurotrophic factor across the blood-brain barrier. In *Neuropharmacology* (Vol. 37).
- Rantamäki, T., Vesa, L., Antila, H., Lieto, A., Tammela, P., Schmitt, A., Lesch, K. P., Rios, M., & Castrén, E. (2011). Antidepressant drugs transactivate trkb neurotrophin receptors in the adult rodent brain independently of bdnf and monoamine transporter blockade. *PLoS ONE*, *6*(6). <https://doi.org/10.1371/journal.pone.0020567>
- Roos, R. A. C. (2010). Huntington's disease: A clinical review. In *Orphanet Journal of Rare Diseases* (Vol. 5, Issue 1). <https://doi.org/10.1186/1750-1172-5-40>
- Rubinsztein, D. C., Leggo, J., Coles, R., Almqvist, E., Biancalana, V., Cassiman, J.-J., Chotai, K., Connarty, M., Craufurd, D., Curtis, A., Curtis, D., Davidson, M. J., Differ, A.-M., Dode, C., Sherr, M., Abbott, M. H., Franz, M. L., Graham, C. A., Harper, P. S., ... Hayden, M. R. (1996). Phenotypic Characterization

- of Individuals with 30-40 CAG Repeats in the Huntington Disease (HD) Gene Reveals HD Cases with 36 Repeats and Apparently Normal Elderly Individuals with 36-39 Repeats. In *Am. J. Hum. Genet* (Vol. 59).
- Sahay, A. S., Sundrani, D. P., & Joshi, S. R. (2017). Neurotrophins: Role in Placental Growth and Development. *Vitamins and Hormones*, 104, 243–261. <https://doi.org/10.1016/BS.VH.2016.11.002>
- Schlessinger, J. (2000). Cell Signaling by Receptor Tyrosine Kinases (receptor) are monomers in the cell membrane. Ligand binding induces dimerization of these receptors resulting in autophosphorylation of their cytoplasmic domain. In *Cell* (Vol. 103).
- Shacham, T., Sharma, N., & Lederkremer, G. Z. (2019). Protein misfolding and ER stress in Huntington's disease. In *Frontiers in Molecular Biosciences* (Vol. 6, Issue APR). Frontiers Media S.A. <https://doi.org/10.3389/fmolb.2019.00020>
- Simmons, D. A. (2017). Modulating Neurotrophin Receptor Signaling as a Therapeutic Strategy for Huntington's Disease. In *Journal of Huntington's Disease* (Vol. 6, Issue 4, pp. 303–325). IOS Press. <https://doi.org/10.3233/JHD-170275>
- Simmons, D. A., Belichenko, N. P., Yang, T., Condon, C., Monbureau, M., Shamloo, M., Jing, D., Massa, S. M., & Longo, F. M. (2013). A small molecule TrkB ligand reduces motor impairment and neuropathology in R6/2 and BACHD mouse models of huntington's disease. *Journal of Neuroscience*, 33(48), 18712–18727. <https://doi.org/10.1523/JNEUROSCI.1310-13.2013>
- Simmons, D. A., Mills, B. D., Butler, R. R., Kuan, J., McHugh, T. L. M., Akers, C., Zhou, J., Syriani, W., Grouban, M., Zeineh, M., & Longo, F. M. (2021). Neuroimaging, Urinary, and Plasma Biomarkers of Treatment Response in Huntington's Disease: Preclinical Evidence with the p75NTR Ligand LM11A-31. *Neurotherapeutics*, 18(2), 1039–1063. <https://doi.org/10.1007/s13311-021-01023-8>
- Soderquist, R. G., Milligan, E. D., Sloane, E. M., Harrison, J. A., Douvas, K. K., Potter, J. M., Hughes, T. S., Chavez, R. A., Johnson, K., Watkins, L. R., & Mahoney, M. J. (2009). PEGylation of brain-derived neurotrophic factor for preserved biological activity and enhanced spinal cord distribution. *Journal of Biomedical Materials Research - Part A*, 91(3), 719–729. <https://doi.org/10.1002/jbm.a.32254>
- Staats, P. S. (2008). Complications of intrathecal therapy. *Pain Medicine*, 9(SUPPL. 1). <https://doi.org/10.1111/j.1526-4637.2008.00445.x>
- Study of LM11A-31-BHS in Mild-moderate AD Patients*. (2017). U.S. National Library of Medicine. <https://clinicaltrials.gov/ct2/show/NCT03069014>
- Tabrizi, S. J., Flower, M. D., Ross, C. A., & Wild, E. J. (2020). Huntington disease: new insights into molecular pathogenesis and therapeutic opportunities. In *Nature Reviews Neurology* (Vol. 16, Issue 10, pp. 529–546). Nature Research. <https://doi.org/10.1038/s41582-020-0389-4>
- Tao, Y. S., Piao, S. G., Jin, Y. S., Jin, J. Z., Zheng, H. L., Zhao, H. Y., Lim, S. W., Yang, C. W., & Li, C. (2018). Expression of brain-derived neurotrophic factor in kidneys from normal and cyclosporine-treated rats. *BMC Nephrology*, 19(1). <https://doi.org/10.1186/s12882-018-0852-2>
- Todd, D., Gowers, I., Dowler, S. J., Wall, M. D., McAllister, G., Fischer, D. F., Dijkstra, S., Fratantoni, S. A., van de Bospoort, R., Veenman-Koepke, J., Flynn, G., Arjomand, J., Dominguez, C., Munoz-Sanjuan, I., Wityak, J., & Bard, J. A. (2014). A monoclonal antibody TrkB receptor agonist as a potential therapeutic for huntington's disease. *PLoS ONE*, 9(2). <https://doi.org/10.1371/journal.pone.0087923>
- Zhang, X., Gureasko, J., Shen, K., Cole, P. A., & Kuriyan, J. (2006). An Allosteric Mechanism for Activation of the Kinase Domain of Epidermal Growth Factor Receptor. *Cell*, 125(6), 1137–1149. <https://doi.org/10.1016/j.cell.2006.05.013>
- Zhao, X., Chen, X. Q., Han, E., Hu, Y., Paik, P., Ding, Z., Overman, J., Lau, A. L., Shahmoradian, S. H., Chiu, W., Thompson, L. M., Wu, C., & Mobley, W. C. (2016). TRiC subunits enhance BDNF axonal transport and rescue striatal atrophy in Huntington's disease. *Proceedings of the National Academy of Sciences*, 113(12), 3233–3238. <https://doi.org/10.1073/pnas.1518111113>

Sciences of the United States of America, 113(38), E5655–E5664.

<https://doi.org/10.1073/pnas.1603020113>

Zhou, H., Wang, J., Jiang, J., Stavrovskaya, I. G., Li, M., Li, W., Wu, Q., Zhang, X., Luo, C., Zhou, S., Sirianni, A. C., Sarkar, S., Kristal, B. S., Friedlander, R. M., & Wang, X. (2014). N-acetyl-serotonin offers neuroprotection through inhibiting mitochondrial death pathways and autophagic activation in experimental models of ischemic injury. *Journal of Neuroscience*, 34(8), 2967–2978.

<https://doi.org/10.1523/JNEUROSCI.1948-13.2014>

Zuccato, C., & Cattaneo, E. (2007). Role of brain-derived neurotrophic factor in Huntington's disease. In *Progress in Neurobiology* (Vol. 81, Issues 5–6, pp. 294–330).

<https://doi.org/10.1016/j.pneurobio.2007.01.003>

Zuccato, C., Ciammola, A., Rigamonti, D., Leavitt, B. R., Goffredo, D., Conti, L., MacDonald, M. E., Friedlander, R. M., Silani, V., Hayden, M. R., Timmusk, T., Sipione, S., & Cattaneo, E. (n.d.). *Loss of huntingtin-mediated BDNF gene transcription in Huntington's disease*.

Zuccato, C., Marullo, M., Conforti, P., MacDonald, M. E., Tartari, M., & Cattaneo, E. (2008). Systematic assessment of BDNF and its receptor levels in human cortices affected by Huntington's disease. *Brain Pathology*, 18(2), 225–238. <https://doi.org/10.1111/j.1750-3639.2007.00111.x>