Analysis of the effectiveness and risk benefit of N acetyl cysteine compared to ceftriaxone in the expression of GLT1 transporters

Análisis de la efectividad y riesgo beneficio de la N-acetilcisteína frente a ceftriaxona en la expresión de transportadores GLT1

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Abstract

The concentration of glutamate in the synaptic cleft is regulated primarily by glutamate transporters, especially by the glial glutamate-specific transporter type 1 (GLT-1). Ceftriaxone (CTX), a β-lactam antibiotic, has been reported to significantly increase GLT-1 expression. N-acetylcysteine as a derivative of cysteine, is oxidized into cystine within the brain, increasing the availability of cystine for the glial cystine-glutamate exchanger, this action increases the amount of glutamate exchanged by glial cells, raising the concentration of glutamate within the extra-synaptic space and effectively promoting GLT-1 transcription. It is suspected that the β-lactam antibiotic ceftriaxone is more effective than N-acetylcysteine in upregulating GLT-1 expression. We conducted a systematic review to investigate the effectiveness of the drugs N-acetylcysteine and ceftriaxone with respect to their effect on increasing the expression of GLT-1 transporters in experimental studies. This systematic review was carried out with methodology in accordance with the Cochrane Handbook and reporting consistent with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA). The main objective of this work is to determine the difference in effectiveness of N-acetylcysteine compared to Ceftriaxone in the expression of the GLT-1 transporter. A clear superiority is shown by ceftriaxone, because by itself it can induce an increase in the levels of these proteins either in circumstances where the glutamatergic flux is affected or in control groups, in contrast to N-acetylcysteine. which improves the expression of these transporters only when there is a deficit in the levels of GLT-1.

Keywords: Ceftriaxone, GLT-1, N-Acetylcysteine, Expression

Resumen

La concentración de glutamato en la hendidura sináptica está regulada principalmente por transportadores de glutamato, especialmente por el transportador glial específico de glutamato tipo 1 (GLT-1). Se ha descrito que la ceftriaxona (CTX), un antibiótico β-lactámico, aumenta significativamente la expresión de GLT-1. La N-acetilcisteína, un derivado de la cisteína, se oxida a cistina en el cerebro, lo que aumenta la disponibilidad de cistina para el intercambiador glial cistina-glutamato. Esta acción incrementa la cantidad de glutamato intercambiado por las células gliales, elevando la concentración de glutamato en el espacio extrasináptico y promoviendo eficazmente la transcripción de GLT-1. Se sospecha que el antibiótico β-lactámico ceftriaxona es más eficaz que la N-acetilcisteína para aumentar la expresión de GLT-1. Se realizó una revisión sistemática para investigar la eficacia de los fármacos N-acetilcisteína y ceftriaxona en relación con su efecto sobre el aumento de la expresión de los transportadores GLT-1 en estudios experimentales. Esta revisión sistemática se llevó a cabo con una metodología acorde con el Manual Cochrane y un informe consistente con los Elementos de Informe Preferidos para Revisiones Sistemáticas y Metaanálisis (PRISMA). El objetivo principal de este trabajo es determinar la diferencia en la eficacia de la N-acetilcisteína en comparación con la ceftriaxona en la expresión del transportador GLT-1. La ceftriaxona muestra una clara superioridad, ya que por sí sola puede inducir un aumento en los niveles de estas proteínas, tanto en circunstancias donde el flujo glutamatérgico se ve afectado como en grupos control, a diferencia de la N-acetilcisteína, que mejora la expresión de estos transportadores solo cuando existe un déficit en los niveles de GLT-1.

Palabras clave: Ceftriaxona, GLT-1, N-acetilcisteína, Expresión

Introduction

There is a vast amount of information about glutamatergic transporters and their usefulness in the regulation of neurological disorders. Likewise, the effectiveness of certain drugs such as N-acetylcysteine and ceftriaxone in the treatment of conditions where the GLT-1 transporter is altered and its function decreases is known; but there are no reviews that help to concentrate this information and update knowledge, causing a delay in therapeutic decision-making in research projects or the neurological interaction of said drugs

The present work is a systematic review that aims to truthfully and up-to-date report the results on the role of N-acetyl cysteine and ceftriaxone with respect to their effectiveness in the expression of type 1 glutamate-specific glial transporters.

The objectives of the paper presented here seek to determine the difference in effectiveness of N-acetylcysteine compared to ceftriaxone in the expression of the GLT-1 transporter and, in addition, to evaluate if there are other risks or benefits that can be attributed to it and to be able to include this knowledge in future research.

Thus, the review is divided into 4 chapters, starting from this introduction. The second chapter is the starting point to understand the importance of the glutamate transporter and regulation at the intra and extracellular level, as well as its role in learning and memory. All this allows us to understand the reason for the regulation of its concentration and reuptake, since, if glutamate reuptake fails, many metabolic pathways in both astrocytes and neurons would be compromised, as well as the hyperexcitation and toxicity derived from high levels of extracellular glutamate.

Being an analytical observational study, it will allow an association to be made on the effectiveness of the drugs N-acetylcysteine and ceftriaxone with respect to their effect on the increase in the expression of GLT-1 transporters, in different pathologies carried out in experimental studies. In this way, this review will provide current knowledge that contributes to speeding up decision-making on the use of such drugs in future preclinical research.

Since just a few years ago, the interest in creating new lines of research on specific glial transporters of glutamate type 1 as a therapeutic target has become progressively more evident due to the promising effects of drugs such as N-acetyl cysteine and ceftriaxone, the which have been used to demonstrate their usefulness in increasing the expression of GLT1 transporters and their benefits in scenarios such as pain treatment, psychiatric disorders, drug addiction, etc.

In 2019, Sophie Lebourgeois, María Carmen González-Marín, Johann Antol, Mickael Naassila and Catherine Vilpoux conducted an experimental study in rats in which they sought to assess whether NAC could reduce ethanol self-administration, ethanol-seeking behavior , motivation and reacquisition of ethanol self-administration after withdrawal in ethanol-dependent rats, successfully demonstrating that in dependent rats, low-dose NAC reduced ethanol self-administration and motivation to consume ethanol, evaluated in a paradigm progressive proportion.

An important experimental work was carried out by Erik J. García, David L. Arndt and Mary E. Cain in 2019 where it was studied how CTX could influence rats under various forms of amphetamine consumption, the study managed to report a large amount of results, however, among the most notable is the fact that CTX increases the expression of GLT1 in astrocytes, but not in the nucleus accumbens.

Weronika Krzyżanowska et al. In 2017 they carried out an experimental study in rats with the intention of testing their hypothesis about brain tolerance to ischemia induced by ceftriaxone and NAC, in this study they

were able to demonstrate the expression of both GLT1 transporters in astrocytes and the expression of xCT in all cells subjected to the study

The most recent review on N acetyl cysteine prepared by Dr. Marco Antonio Nocito Echevarria Et Al. (Tassio Andrade Reis, Giulianno Ruffo Capatti, Victor Siciliano Soares, Dartiu Xavier da Silveira, Thiago Marques Fidalgo) in 2017 aimed to review the literature available to date on the use of NAC for cocaine dependence. Among their results, it can be found that NAC reverses the alteration of glutamate homeostasis caused by prolonged cocaine use, restoring the function of the cystine-glutamate exchanger in glial cells and reversing the negatively regulated GLT-1 receptor.

Among the most notable antecedents of recent years we have the work of scientists Douglas J. Roberts-Wolfe and Peter W. Kalivas dating from 2015 and whose purpose was to highlight the effects of the use of addictive drugs on the uptake of GLT-1 and glutamate, and the use of GLT-1 as a target in addiction pharmacotherapy. Among the authors' conclusions is that most of the preclinical and clinical research reviewed in their research suggests that upregulation of GLT-1 is a promising strategy for treating substance use disorders and other disorders characterized in part by compulsive behavior.

A group of Mexican researchers published a study in 2018 where they sought to determine the analgesic and anti-inflammatory effects of the administration of ceftriaxone and clavulanic acid in a model of inflammatory pain induced by Carr in an acute period of time. Their research builds on insights generated by Rothstein et al and succeeded in providing evidence that both CTX and CA cause an analgesic effect in a Carr model of inflammatory pain.

Positive regulators of glutamate type 1 expression (GLT1) can be used in the prevention and/or treatment of neurological disorders (Neha Soni, 2014), due to the activity carried out by the excitatory amino acid

transporter with the clearance mechanism with which it maintains the level of glutamate in the synapse below excitotoxic levels, thus avoiding dysfunctional synaptic interactions in neurodegenerative disorders.

There are several studies in which the use of N-acetyl cysteine and ceftriaxone have been shown to increase the expression of specific glial transporters GLT-1 in the face of induced downregulation, resulting in a range of reported effects under certain circumstances. such as tolerance to ischemia in a stroke (Weronika K, et al 2017), prevention of relapses and coadjuvant in the treatment of drug addiction (Lori A. Knackstedt et al. 2010), intervention in the treatment not only of diseases neurodegenerative diseases such as Huntington's disease (Sari, Y. et al 2010) and pain in diabetic neuropathy (Serena Notartomaso et al 2019)

However, there are still not enough reviews that compile the results of articles on the regulation of glutamate via the increase of GLT-1 transporters with the use of the mucolytic N-acetylcysteine and the beta-lactam antibiotic ceftriaxone and compare the effectiveness between both pharmacological agents in virtue of the mechanism of action and the areas of the central nervous system in which they act.

There is a considerable amount of information on the reported effects of the relationship of N-acetylcysteine and ceftriaxone with the positive regulation of GLT-1, but there are no reviews that help to condense this information and update knowledge, which causes a delay in taking of decisions on the choice of the use of these drugs in research projects or there may even be ignorance of the neurological interaction of these drugs, reducing the possibility of evaluating whether there are other risks or benefits that can be attributed to them and being able to include this knowledge in future studies. investigations thus reducing the possibility of the joint use of these pharmacological agents with neuroprotective effects with the current treatment of these neurological disorders such as epilepsy, Huntington's disease, tolerance to ischemia in stroke or psychosis in humans.

Heterogeneity can be a difficulty considering that N-acetylcysteine and ceftriaxone belong to different pharmacological categories according to the ATC code (María Verónica S., 2004) of anatomical-therapeutic-chemical classification of drugs, N-acetylcysteine to the respiratory system group, preparations for cough and cold, with a mucolytic mechanism of action and ceftriaxone is from the group of systemic anti-infectives, a broad-spectrum cephalosporin that inhibits cell wall synthesis. However, the ability to regulate glutamate that has been reported in both drugs has prompted this project to answer the following. Which drug has greater efficacy when comparing ceftriaxone and N-acetylcysteine in the expression of the GLT-1 transporter?

The alteration of glutamatergic transmission is associated with cognitive and motor alterations of various pathologies. If glutamate reuptake fails, in addition to the hyperexcitation and toxicity derived from high levels of extracellular glutamate, many metabolic pathways in both astrocytes and neurons would be compromised, including those that allow synthesizing neurotransmitters in excitatory and inhibitory neurons.

Several studies have shown that the use of N-acetylcysteine and ceftriaxone increase the expression of glial-specific glutamate transporters type 1 (GLT-1), resulting in a range of effects reported under certain circumstances, mentioned above.

β-lactam antibiotics, including ceftriaxone (CEF), upregulate GLT-1 expression and have neuroprotective effects (Rothstein et al. 2005).

This research work is highly relevant since it will allow comparing the effect of N-acetylcysteine and ceftriaxone in the different conditions where the GLT-1 transporter is altered and its function decreases; as in neurodegenerative diseases, drug addiction and psychiatric disorders. It will also make it possible to compare its usefulness in the treatment of these conditions and to assess whether there are other risks or benefits that can be attributed to it and to be able to include this knowledge in future research.

Glutamate.

Glutamate is an amino acid with great importance in brain metabolism that also acts as the main excitatory neurotransmitter in the brain. Glutamate participates directly and indirectly in a wide variety of brain functions. It plays a very important role in the pathogenesis of many neurological disorders such as epilepsy, ALS (amyotrophic lateral sclerosis), cerebral ischemia, schizophrenia, parkinsonism, and Alzheimer's disease (Nakagawa and Kaneko, 2013; Annweiler et al., 2014; Kleteckova et al. ., 2014).

Its release in the synaptic space by glutamatergic neurons allows its binding to postsynaptic receptors, favoring the depolarization of the postsynaptic neuron. In this way, a new electrical impulse is generated in the postsynaptic neuron.

Glutamate transporters.

Glutamate transporters are membrane-bound proteins located on glial cells and/or presynaptic glutamatergic nerve endings and are essential for the removal and termination of action of the excitatory neurotransmitter glutamate from the synapse (Shashidharaan et al., 1994).

There are several families of glutamate transporters, and they include the plasma membrane excitatory amino acid transporters (EAATs), the vesicular glutamate transporters (VGLUTs), and the glutamate-cysteine exchanger (Anderson and Swanson, 2000). Five types of glutamate transporters have been described: EAAT1 to EAAT5, present in glial cells (EAAT1 and EAAT2), in neurons (EAAT3 and EAAT4) and in the retina (EAAT5).

GLT-1/EAAT2 is found primarily responsible for maintaining glutamate homeostasis, which if disturbed due to downregulation of these transporters causes excitability which in turn leads to neurodegeneration.

Glutamate transporters couple glutamate uptake with inorganic ion transport, thus using stored free energy as electrochemical potential gradients of these ions to drive uphill transport (Grewer et al., 2000). This

coupling mechanism is essential for the efficient removal of glutamate from extracellular spaces such as the cerebrospinal fluid, the intestinal lumen, and the lumen of the renal proximal tubules.

N acetylcysteine

N-acetylcysteine (NAC) is a derivative of the amino acid cysteine with an acetyl group attached to nitrogen, which was developed as a mucolytic in the 1960s, it can act as a precursor or reduced glutathione and as a direct scavenger of reactive oxygen species. (Prabhu et al., 2009; Gillissen., 2011).

NAC produces repletion of glutathione stores, which confers antioxidant activity, and the thiol portion of the molecule confers direct antioxidant properties. (Prabhu et al., 2009). The use of NAC in the treatment of chronic acute bronchitis has also been demonstrated. In humans, NAC has been shown to improve idiopathic pulmonary fibrosis, various forms of alveolitis, and prevent the hepatotoxic effects of paracetamol (Gillessen., 2011). Its action as an antioxidant is little known and, possibly, NAC is one of the most powerful antioxidant molecules to which, of course, a beneficial therapeutic application can be attributed (Garcia et al., 2020).

Ceftriaxone

Although beta-lactams have historically been used as antimicrobials, a notable side effect on the host has been identified (Rothstein et al. 2005). Beta-lactam antibiotics, such as ceftriaxone, have been shown to enhance ex vivo expression of a neuroprotective protein GLT-1/EAAT-2 in a concentration-dependent manner (Karaman et al., 2013). GLT-1/EAAT-2 terminates the potentially neurotoxic effects of the neurotransmitter glutamate by removing it from the synaptic cleft.

Beta-lactam-mediated activation of GLT-1/EAAT-2 expression is proposed to involve nuclear factor-kB (NF-kB), whereby β-lactamases induce nuclear activation of this transcription factor (Feng et al. al., 2014). Activated NF-kB then binds to the GLT-1/EAAT-2 promoter region and upregulates transcription of this gene

(Lee et al., 2008), thus decreasing the concentration of glutamate in the synaptic cleft and alleviating the effects potentially neurotoxic effects of excess glutamate (Salles et al., 2014).

Some studies also found that ceftriaxone treatment can attenuate neuronal injury and improve spatial learning and memory after chronic cerebral hypoperfusion and that glutamate excitotoxicity may play an important role in the pathophysiology of chronic cerebral hypoperfusion (Koomhin et al., 2012). This ceftriaxone-induced upregulation of GLT-1/EAAT-2 blocks metabotropic glutamate receptor (mGluR)-dependent long-term depression (LTD) in the mossofiber (MF)-capped hippocampal CA3 synapse. It also has negative effects on long-term potentiation (LTP).

Ceftriaxone is currently in human clinical trials for ALS (Zhao et al., 2014). The transition from preclinical studies in mice to clinical trials in humans is very difficult; Successful preclinical studies often fail in clinical trials.

The role that GLT-1 may have in pathophysiological conditions in neurons is not fully understood, although there is evidence that, under certain conditions, neuronal GLT-1 may be important. It has been described that neuronal GLT-1 may be increased when there is a deficiency of glutamate transport by astrocytes (Pow et al., 2004; Rimmele and Rosenberg, 2016). Certain studies show that, under conditions of stress or brain damage, proteins typically expressed in astrocytes could become expressed in neurons in an attempt to compensate for the lack of glutamate uptake.

Among these proteins is GLT-1, whose increased expression in neurons has already been described in diseases such as Alzheimer's (Pow and Cook, 2009; Thal, 2002) or cerebral hypoxia (Pow et al., 2004). For this reason, an increase in the expression of neuronal GLT-1 in damage conditions is proposed with a protective role that prevents neurotoxicity.

However, having an overexpression of GLT-1 has also been associated with neuronal damage and neuronal death. In 2005, Selkirk et al published a study where, in organotypic cultures of mouse hippocampus, GLT-1 was specifically overexpressed in neurons and subjected to acute concentrations of glutamate.

The explanation offered by the scientific community for this apparent contradiction is that GLT-1 is probably expressed in neurons in response to damage and an increase in glutamate in the neuron. However, in the presence of continued damage, this apparent protection ceases to be such and begins to have deleterious effects for the cell, favoring its death by excitotoxicity.

Methodology

We conducted a systematic review and meta-analysis to investigate the effectiveness of the drugs N-acetylcysteine and ceftriaxone with respect to their effect on increasing the expression of GLT-1 transporters in experimental studies with rats. II The articles eligible for inclusion were descriptive and analytical studies of an experimental or observational type without distinction in their temporal sequence or their temporal relationship study factor – disease, excluding those studies that do not have an evaluation of bias equal to or less than low risk. . This systematic review was carried out with methodology in accordance with the Cochrane Handbook and reporting consistent with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA).

Search strategy

We designed a high-sensitivity search strategy that combines groups of synonyms of free-text search terms and keywords for GLT1, combined with groups for ceftriaxone, N-acetylcysteine, and expression. In the next step we systematically search for articles published in MEDLINE, PUBMED, MEDSCAPE, Open Access Journals and Google Scholar. Additional searches of preprint servers (Biorxiv, Medrxiv, and Chinxiv) using

keywords “Expression, ceftriaxone, N-acetylcysteine” were performed to identify potential prepublished manuscripts that met the eligibility criteria. All of these searches spanned from January 2008 to July 31, 2021.

For greater sensitivity, we then performed a second expanded search of the PUBMED, MEDLINE, and Open Access Journals databases from January 1, 2014, through July 31, 2021, for all published meta-analyses and reviews reporting the expression of GLT-1 by ceftriaxone and n-acetylcysteine without distinction in the therapeutic / clinical effect implied in the studies.

No exclusions were made by language or species studied. Search results from the used databases as well as preprint servers were manually scanned to identify eligible studies. Reference lists of all included articles were also checked for possible citation eligibility.

Study selection and data extraction

Studies were examined using titles and abstracts followed by full text review. Studies were included if they were randomized clinical trials, cohort studies, reviews, or meta-analyses describing a population of species dosed with n-acetylcysteine or ceftriaxone and reporting their effect on GLT-1 transporters.

Data extraction was performed using a standardized data analysis table and the data included in it, from the text, tables and graphs of the article.

A total of 70 articles available from January 2008 to July 2021 were analyzed, 32 were discarded due to repetition or did not have the relevant data to enrich the present work.

Conclusions

NAC

N-acetyl cysteine has been used for many years as a mucolytic agent, however, lines of research for some decades have suggested its use for the treatment of drug addiction, neuropsychiatric disorders, pain, among

others. As a derivative of cysteine, it is oxidized to cystine within the CNS, increasing the availability of cystine for the glial cystine-glutamate exchanger. This action increases the amount of glutamate exchanged by glial cells, raising the concentration of glutamate within the extrasynaptic space and effectively restoring GLT-1 transporters (MA et al., 2017).

In the present work, the role of NAC in the expression of glial-specific glutamate transporters type 1 (GLT-1) is reviewed; A total of (9) articles were analyzed, of which (7) were experimental studies, (2) review articles, and (1) meta-analysis.

The data show that N acetyl cysteine has the ability to increase the number of GLT-1 transporters, however, in (6) studies emphasize that for this effect to be achieved there must be a deregulation of glutamatergic flow, otherwise NAC has no effect on transporters. (Quintanilla et al., 2016)

In 2019, a study was carried out in which an attempt was made to restore the GLT-1 transporters that had decreased on purpose through the chronic consumption of ethanol in adult female Wistar rats, the results reported that NAC did cause a significant increase in the number of transporters in the glial cells of the hippocampus and the prefrontal cortex but it was not enough to normalize the levels of GLT-1, however, when administering NAC+ASA at doses of 40 mg/kg and 15 mg/kg, respectively, normalization of the transporters was achieved and glutamatergic flux. (Israel et al., 2019).

CTX

Ceftriaxone is a beta-lactam antibiotic belonging to the third-generation cephalosporins that has been shown in recent years to be useful in many disorders of the nervous system by inhibiting excitotoxicity through the upregulation of astrocytic GLT-1. It has been reported that it can induce transcription of the GLT-1 gene by activating nuclear factor kappa light chain enhancer of activated B cells (NF-kB) (Lee et al., 2008).

Here we present the information available to date on the usefulness of ceftriaxone to increase the transcription of GLT-1 transporters.

A total of 22 studies showed that ceftriaxone is significantly useful for increasing the transcription of the glutamate-specific glial transporter type 1, which has allowed it to be included in studies where there are abnormalities in its expression. One point in favor of this drug is that its effect has a place in various locations of the central nervous system, since it has been shown that intraperitoneal injections at doses of 200 mg/kg achieved a significant increase in at least three locations of high importance in the central nervous system (see later results according to the expression zones) detected by Western Blot. (Wilkie et al., 2021)

Another aspect that can be attributed to ceftriaxone is its use in the field of cognitive impairment, since its administration relieves symptoms by improving glutamate uptake by GLT-1; however, chronic administration of ceftriaxone induced a significant impairment of memory in object recognition tests. Furthermore, it disrupted motor skill learning and functional outcome after focal ischemic cortical injury (Gao et al., 2020), thus ceftriaxone use in this field is reserved for acute administration.

In general, regarding the effectiveness between these two drugs to achieve an increase in the transcription of GLT-1 transporters, a clear superiority is shown by ceftriaxone, because by itself it can induce an increase in the levels of said proteins either in circumstances where the glutamatergic flow is affected or in control groups, in contrast, N acetyl cysteine improves the expression only when there is some abnormality in the levels of GLT-1 (eg in drug addicts it has been shown that the chronic consumption of these substances reduces the number of transporters in the membranes of astrocytes), and even in these circumstances at a dose of 150 mg/kg/day, its effect may not be sufficient to normalize the amount of these proteins in the membranes, however, it has been shown that this pharmacological property can be enhanced by other drugs such as acetylsalicylic acid, restoring and even improving the expression of transpo GLT1 receptors;

However, in control groups where no alteration in the glutamatergic cycle is found, NAC induces a reduction in GLT-1 transporters, possibly due to an immunomodulatory effect mediated by NF-kB.

Effectiveness according to the areas in which regulation of Glt1 expression has been reported with ceftriaxone and N-acetylcysteine.

GLT-1 is a sodium-dependent excitatory amino acid transporter type 2 (EAAT2) protein that plays an important role in the removal of glutamate after its release into the extracellular space. The high-affinity transporter GLT-1 is abundantly localized to astrocytes and axon terminals in some parts of the mature brain (Xiao Luo et al. 2020); and is a fundamental part of the glutamate-glutamine cycle necessary for glutamate production in glutamatergic neurons, and reuptake in astrocytes. This transporter protein represents more than 1% of the total tissue protein in the hippocampus (Lehre and Danbolt, 1998).

The upward modulations of Glt1 after the application of ceftriaxone have been described according to the articles reviewed, in different areas of the nervous system after induced decreases of this protein, it has been shown that the hippocampus is the area where there is more regulation produced by ceftriaxone at a standard dose of ceftriaxone administered in rats of 200 mg/kg, in more than 45% (11) of reviewed ceftriaxone studies, in (29%) (7) it is also the prefrontal cortex, to a lesser extent quantity was positively expressed in the striatum (Sari, Y., Prieto, AL, Barton, SJ et al,1010) and nucleus accumbens (Weronika K, et al. 2017).

In studies of neuropathic pain, the upregulation of Glt1 upon administration of ceftriaxone by different routes, in an interval of between 5 and 8 days, has shown an increase in the dorsal horn of the spinal cord and the periaqueductal gray matter (Zhuoyang Zhao et al. 2018). Up to 77% increases in dimerized membrane-bound

GLT-1 protein expression were found in the lumbar spinal cord with ceftriaxone administered intrathecally once daily (K. M. RAMOS et al 2010)

We should also add that some studies report decreased basal glutamate levels in the frontal cortex and hippocampus (Irene Smaga 2020, Crystal M et al, 2021) and suggest that ceftriaxone may alter glutamate release properties independently of its effects on GLT-1 expression.

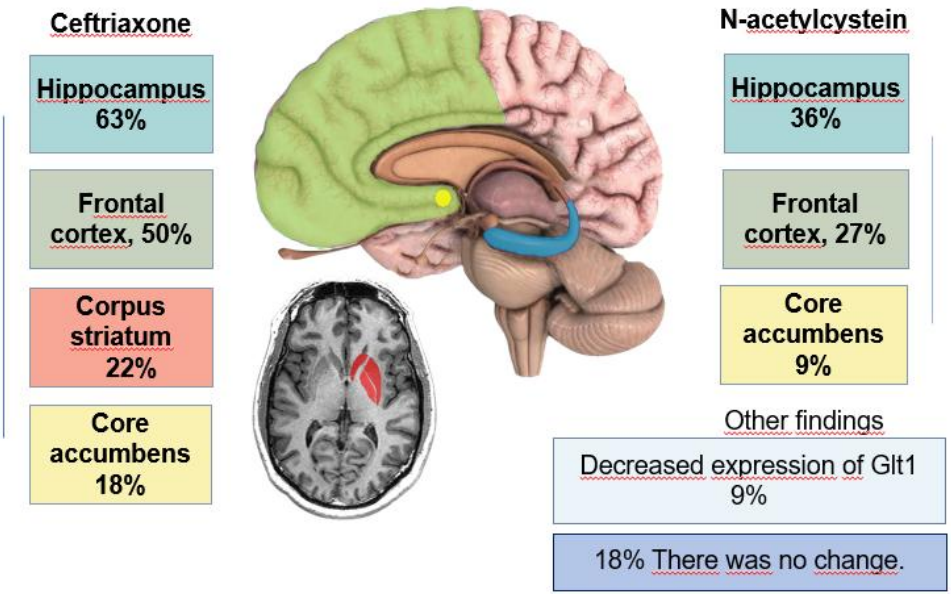
Unlike the areas of the nervous system in which Glt1 regulations are reported after the application of N-acetylcysteine, where it has been shown that this transporter protein restores GLT1, however, the mechanism of action is not yet known. Although lack of change in deficit in induced GLT-1, in the striatum and cortex were detected in 33% of the reviewed studies of N-acetylcysteine (Serena N, et al 2019), and in more recent studies regulation results are evidenced downregulation of GLT1 expression due to the possible inhibition of the NF - κB pathway, which is a bidirectional regulator of GLT1. (Mark D. Namba et al, 2019). Another theory is that GLT-1 dependent glutamate uptake is hampered by the presence of a high extracellular glutamate to cysteine ratio which may explain why GLT-1 is decreased in Huntington's disease.

It is also worth noting the presence of glutamate regulation in the nucleus accumbens due to the use of N-acetyl cysteine, however, associated with the GLT1-independent cysteine-glutamate transporter.

Ceftriaxone demonstrates a clear superiority in the effectiveness for the expression of GLT1 transporters both in groups with impaired glutamatergic flux regulated down and in control groups, significantly increasing these proteins in the nucleus accumbens, striatum, frontal cortex and hippocampus.

The effectiveness of N acetyl cysteine with these transporters takes place only in cases where there is a deregulation in the glutamate cycle, being able to achieve normalization of GLT1 levels in the hippocampus, nucleus accumbens and frontal cortex; If there is no alteration in said cycle, NAC can cause a downregulation of these transporters due to its immunomodulatory mechanism mediated by NF-kB.

Figure 1. Percentage of upward modulation of GLT-1 reported in different áreas of the nervous system



The effect of NAC in normalizing GLT1 levels may be potentiated by ASA.

There is a possibility that when ceftriaxone and n-acetyl cysteine are administered together at doses of 200 mg/kg and 150 mg/kg intraperitoneally, there is a potentiation by the mechanism mediated by NF-kB.

Discussion

The hypothesis of the present study was that there would possibly be a superiority of ceftriaxone over n-acetyl cysteine on the expression of type 1 glutamate glial transporters. studies show findings that guide the benefits of ceftriaxone on NAC in the role of GLT1 transcription, for example, Roberts Wolfe and Kalivas compiled information with which they manage to determine that ceftriaxone administered alone can improve the expression of these transporters at various sites in the CNS (eg, nucleus accumbens, hippocampus, hypothalamus, and striatum) either in rat models in which glutamatergic cycle dysregulation has been elicited or in control groups.

In contrast, NAC only shows an upregulation of GLT-1 in groups whose glutamatergic flux has been altered (drug addiction or neuropsychiatric disorders such as Huntington's disease); In addition, it is important to mention that if there is no alteration in the glial transporters of glutamate type 1, it can trigger a downregulation of these proteins mediated by NF-kB. In 2019, Lebourgeois et al. conducted a study with male Wistar rats and demonstrated using the Western blood method that NAC decreased GLT-1 levels by up to 28% compared to a control group. (p 0.739).

Likewise, it seems that both CTX and NAC have an important role in the expression of GLT-1 transporters individually, however, in the study carried out by Krzyzanowska et al., these two drugs were administered separately and in combination at doses of 250 mg/kg and 150 mg/kg for ceftriaxone and n-acetyl cysteine, respectively, showing a clear advantage in the effectiveness of the expression of these transporters by administering the drugs in combination intraperitoneally, causing an increase in GLT-1 in the frontal cortex ($p < 0.001$) and hippocampus (<0.001); However, the results of NAC administered alone showed that it decreases these transporters in the hippocampus and striatum.

With which, and since there is no consistent evidence on the subject, we could propose a new hypothesis about the fact that the mechanism of action of NAC mediated by NF-kB that triggers a downregulation of GLT-1 may be in actually an enhancer for CTX to achieve an even greater increase in these proteins.

Another unanswered finding that is evidenced in this work is the fact that ceftriaxone, when administered to adult male Wistar rats, causes an increase in glial glutamate transporters in the dorsal horn of the spinal cord, frontal cortex and hippocampus, however When the same drug was administered to neonatal rats, an increase in these proteins could only be detected in the cortex, which gives rise to the question: What characteristics do other areas of the CNS have that allow CTX to cause an increase in the number of transporters?

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