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Regulation of neurotransmitter release by endogenous nitric oxide in striatal slices

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Abstract

This study sought to determine the potential role of nitric oxide (NO) in *N*-methyl-D-aspartate (NMDA)-stimulated efflux of [^{14}C] γ-aminobutyric acid (GABA) and [^{3}H]acetylcholine from striatal slices in vitro. In Mg^{2+}-free buffer, NMDA-stimulated [^{14}C]GABA and [^{3}H]acetylcholine release were inhibited by the guanylate cyclase inhibitor, 1*H*-[1,2,4]oxadiazolo[4,3-a]quinoxalin-1-one (ODQ), and, to a lesser extent, by the nitric oxide synthase inhibitor, nitroarginine (N-Arg). Since reversal of catecholamine transporters previously has been implicated in the mechanism underlying NO-induced catecholamine release, we used the GABA transport inhibitor, 1-(2-(((diphenylmethylene)imino)oxy)ethyl)-1,2,5,6-tetrahydro-3-pyridine-carboxylic acid hydrochloride (NNC-711), to address the role of GABA transport in NArg-sensitive NMDA-induced release. NNC-711 inhibited NMDA-stimulated [^{14}C]GABA efflux by 50%, confirming our previous report that NMDA-stimulated GABA release is partially dependent on reversal of the transporter. The effect of N-Arg in the presence of NNC-711 was similar to its effect in the absence of the transport inhibitor, suggesting that reversal of the transporter is not involved in the NO component of NMDA-stimulated [^{14}C]GABA release. These data suggest that glutamatergic transmission through striatal NMDA receptors is partially mediated through activation of the NO/guanylate cyclase pathway and that this mechanism may contribute to the tetrodotoxin sensitivity of NMDA-induced release of GABA and acetylcholine in the striatum.

Keywords: Acetylcholine release; GABA (γ-aminobutyric acid) release; Nitric oxide (NO); NMDA receptor; Striatum

1. Introduction

Nitric oxide (NO) is a neuronal messenger that is thought to play a role in synaptic plasticity. The role of NO in long-term potentiation or long-term depression likely involves an action on transmitter release (Bohme et al., 1991; O'Dell et al., 1991; Schuman and Madison, 1991; Shibuki and Okada, 1991; Boulton et al., 1994). NO or NO donors have been shown to differentially modulate acetylcholine, γ-aminobutyric acid (GABA), glutamate and dopamine release in different brain regions (Lonart et al., 1992, 1993; Prast and Philippu, 1992; Hirsch et al., 1993; Guevara-Guzman et al., 1994; Segovia et al., 1994; Stout and Woodward, 1994; Lonart and Johnson, 1994, 1995a,b; Sandor et al., 1995; Takita et al., 1997). Stimulation of neurotransmitter release in cortex and cerebellum by activation of NMDA receptors has been shown to involve the production of NO (Montague et al., 1994; Okhuma et al., 1995; Oh and McCaslin, 1996). Although activation of NO synthesis by *N*-methyl-D-aspartate (NMDA) has been shown to result in the production of cyclic-3′,5′-guanosine monophosphate (cGMP) in several systems including the striatum (Garthwaite et al., 1988, 1989; Bredt and Snyder, 1989; Southam et al., 1991; East et al., 1996), the role of the NO/cGMP pathway in NMDA-stimulated transmitter release, particularly in the striatum, is unclear (Segovia et al., 1994; Moller et al., 1995; Semba et al., 1995).

The majority of striatal neurons are medium sized, spiny GABAergic interneurons or projection neurons (Angulo and McEwen, 1994). 1–2% of striatal neurons are large aspiny neurons that use ACh as their neurotransmitter. These cholinergic neurons are mostly interneurons, although a subpopulation may project to neocortex (Di Chiara et al., 1994). A subclass of striatal interneurons are medium size, aspiny interneurons in which nitric oxide synthase is colocalized with neuropeptide Y, GABA and

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somatostatin (Albin, 1995; Kawaguchi, 1993, 1997). All of these striatal neurons receive excitatory glutamatergic input from the cortex, and they all express the NMDA receptor (Graybiel, 1990; Landwehrmeyer et al., 1995; Penney et al., 1996). Therefore, in this study we sought to determine the potential role of NO and cGMP in the mechanisms underlying the release of GABA and ACh upon activation of striatal NMDA receptors.

2. Materials and methods

2.1. [^{14}C]GABA and [^{3}H]ACh release

Male Sprague–Dawley rats weighing 175–200 g were used throughout these experiments. After decapitation, the striatum was dissected and placed in ice-cold modified Kreb's-bicarbonate buffer (pH 7.45) containing 118 mM NaCl, 3.3 mM KCl, 1.25 mM $CaCl_2$, 1.2 mM KH_2PO_4, 25 mM $NaHCO_3$, 15.1 mM glucose, 10 mM HEPES, 0.03 mM EDTA and 0.1 mM aminooxyacetic acid (a GABA transaminase inhibitor to prevent GABA metabolism). Striatal cross-chopped slices (400 μm $\times$ 400 μm) were prepared using a McIIwain tissue chopper. Slices were washed three times with ice-cold buffer, then loaded with 100 nM [^{3}H]choline and 4 μM [^{14}C]GABA for 15 min at 34°C and then placed in superfusion chambers. Slices were superfused continuously with oxygenated (95% O_2-5% CO_2) modified Kreb's-bicarbonate buffer at a rate of 0.3 ml/min for 1 h. After equilibration, three 5-min fractions were collected to determine basal [^{14}C]GABA and [^{3}H]ACh efflux. All drugs were introduced 15–25 min prior to NMDA stimulation. At the end of the experiment, radioactivity in each fraction and the radioactivity in the slices were determined using liquid scintillation spectrometry. Fractional ^{3}H and ^{14}C efflux in the 5-min fractions was expressed as the amount of radioactivity in the superfusate relative to the total amount of radioactivity at that particular time point, multiplied by 100. The effect of various drugs on basal and NMDA-stimulated release is expressed as the area under the curve (AUC). In either case, the basal fractions prior to the addition of either test drug or NMDA were averaged to estimate the baseline and this value then was subtracted from each of the subsequent fractions in order to estimate the AUC.

Early in these experiments we observed that the inclusion of hemicholinium-3 to prevent the reuptake of [^{3}H]choline derived from the hydrolysis of recently released [^{3}H]ACh, substantially diminished NMDA-stimulated [^{14}C]GABA efflux. The mechanism of this inhibition was not investigated further, but preliminary experiments comparing NMDA-evoked tritium efflux in the presence and absence of hemicholinium-3 showed only modest differences. After loading striatal slices with [^{3}H]choline, the fractional release of tritium in the absence of hemicholinium-3 was about 50% less under basal conditions as compared to the presence of hemicholinium-3. However, NMDA-stimulated efflux was not significantly affected. Thus, in this paradigm, basal efflux is somewhat underestimated, but stimulated efflux of ACh is not significantly affected by the omission of hemicholinium-3.

Data are presented as means $\pm$ S.E.M. Statistical differences were determined by Student's *t*-test. A probability of $P < 0.05$ was considered significant.

These experiments were carried out in accordance with a protocol that was approved by the Institutional Animal Care and Use Committee of the University of Texas Medical Branch at Galveston. This protocol adheres to the principles set forth by the European Community regarding the use of experimental animals.

2.2. Materials

1-(2-(((diphenylmethylene)imino)oxy)ethyl)-1,2,5,6-tetrahydro-3-pyridine-carboxylic acid hydrochloride (NNC-711 hydrochloride), NMDA was purchased from Research Biochemicals (Natick, MA, USA). Aminooxyacetic acid and N^{ω}-nitro-L-arginine were purchased from Sigma (St. Louis, MO, USA). 1*H*-[1,2,4]oxadiazolo[4,3-a]quinoxalin-1-one (ODQ) was purchased from Tocris Cookson (St. Louis, MO, USA). [^{3}H]Choline (81 Ci/mmol) and [^{14}C]GABA (223 mCi/mmol) were purchased from New England Nuclear (Boston, MA, USA). All remaining chemicals were obtained from Fisher Scientific (Houston, TX, USA) or Sigma.

3. Results

3.1. Effects of the NNC-711 on basal and NMDA-stimulated [^{14}C]GABA and [^{3}H]ACh release

Previous studies of K^+-evoked release of [^{3}H]GABA or NMDA-induced release of endogenous GABA revealed that the effect of aminooxyacetic acid was identical to that of γ-vinyl GABA (Agostinho et al., 1994; Wang et al., 1996), a reportedly more selective GABA transaminase inhibitor (Hammond and Wilder, 1985). Also, in the study from this laboratory similar effects of tetrodotoxin and NNC-711 as reported below were observed (Wang et al., 1996). Thus, the efflux of [^{14}C]GABA under these conditions appears to be a good approximation of endogenous GABA release. Our initial characterization of NMDA-stimulated [^{14}C]GABA and [^{3}H]ACh efflux indicated that both were inhibited by more than 95% by 1.2 mM $MgCl_2$ and 1 μM phencyclidine (data not shown), suggesting that under these conditions, the stimulated efflux of both transmitters is due to NMDA receptor activation.

Superfusion with the GABA uptake inhibitor NNC-711 (20 nM) for 25 min produced a marked increase in basal

[^{14}C]GABA efflux (Fig. 1, upper panel). This effect was blocked by the sodium channel blocker tetrodotoxin (1 μM), suggesting that this increase is the result of diminished reuptake of GABA into spontaneously active neurons. NNC-711 also modestly elevated basal [^{3}H]ACh efflux in a tetrodotoxin-sensitive manner (Fig. 1, lower panel). Preliminary experiments showed that stimulation of striatal slices with NMDA (100 μM–1 mM) for 5 min, induced release of [^{14}C]GABA and [^{3}H]ACh release in a concentration-dependent manner. Subsequent experiments used 300 μM NMDA, a submaximal concentration. NNC-711 (20 nM) significantly inhibited NMDA-stimulated [^{14}C]GABA, but not [^{3}H]ACh release, suggesting either that NMDA stimulates [^{14}C]GABA release partially through reversal of the GABA transporter (Fig. 1, inserts) or that NNC-711 acts indirectly through GABA autoreceptors to inhibit the release of [^{14}C]GABA. However, 30 μM baclofen did not affect [^{14}C]GABA efflux and the $GABA_A$ receptor agonist, isoguvacine, actually potentiated efflux in a picrotoxin-sensitive fashion (data not shown). Thus, both basal and NMDA-stimulated [^{14}C]GABA efflux are tetrodotoxin-sensitive and the latter involves the reversal of the GABA transporter as previously reported for endogenous GABA (Wang et al., 1996).

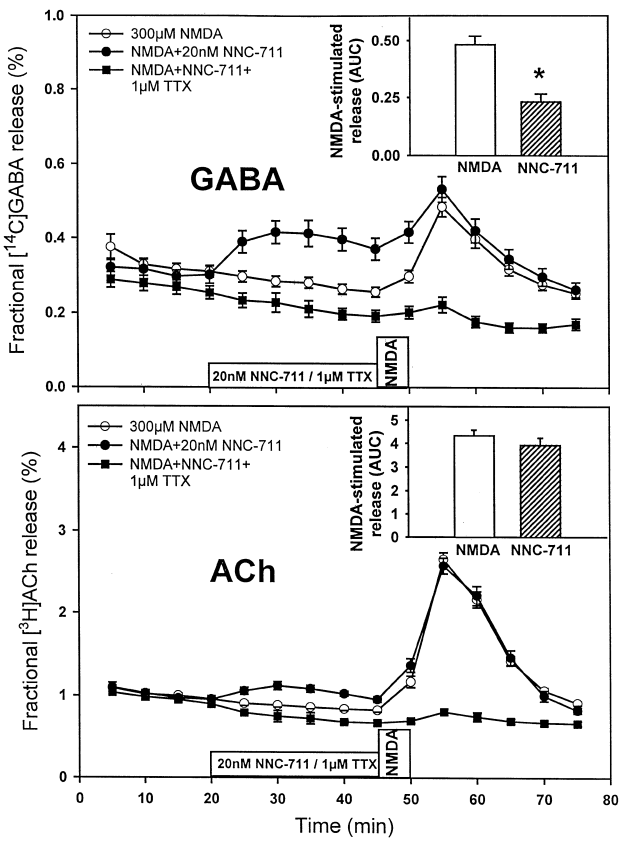

Fig. 1. The effect of NNC-711 and tetrodotoxin (TTX) on basal [^{14}C]GABA and [^{3}H]ACh release from striatal slices. NNC-711 (20 nM) by itself or in combination with 1μM TTX were introduced 25 min prior to stimulation with 300 μM NMDA. NMDA was superfused for 5 min. Data points represent mean ± S.E.M of 22–24 determinations for control (open circle), 8–10 determinations for NNC-711 (closed circle) and 9 determinations for NNC-711 + TTX (closed squares), respectively. The inserts show the effect on NNC-711 on NMDA- stimulated [^{14}C]GABA and [^{3}H]ACh release. Data are presented as mean AUC ± S.E.M. * Significantly different than control, $P < 0.05$.

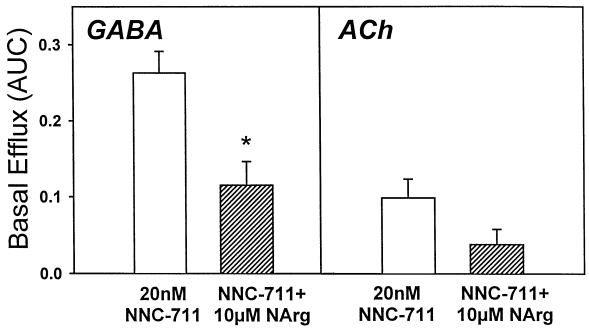

Fig. 2. The role of endogenous NO on NNC-711-stimulated basal [^{14}C]GABA and [^{3}H]ACh efflux from striatal slices. Slices were superfused with 20 nM NNC-711 (open bars, $n = 9–12$) or NNC-711 + 10 μM N-Arg (closed bars, $n = 10$) for 25 min. The data are expressed as mean AUC ± S.E.M. * Significantly different than control, $P < 0.05$.

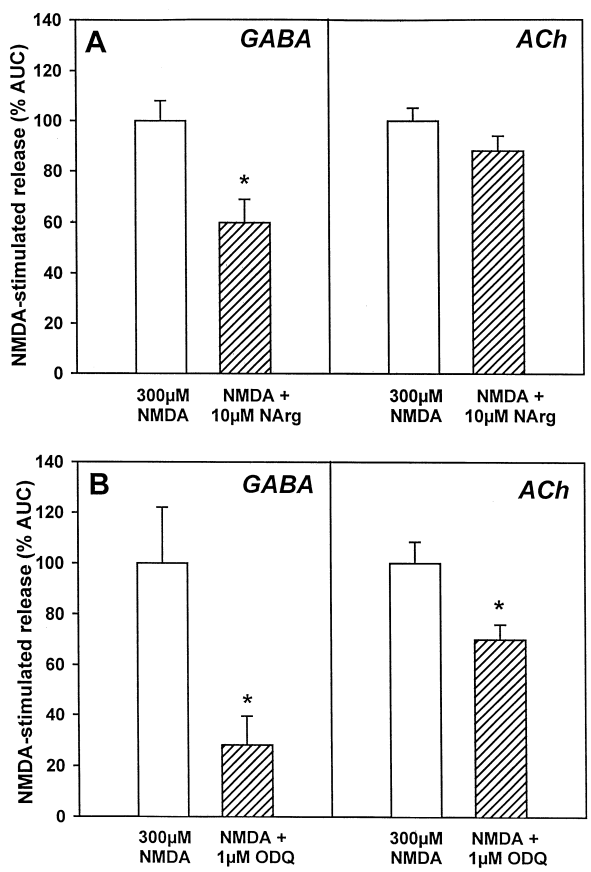

Fig. 3. The role of endogenous NO on NMDA-stimulated [^{14}C]GABA and [^{3}H]ACh release. Striatal slices were superfused with 10 μM N-Arg (A, hatched bars, $n = 14–15$) or 1 μM ODQ (B, hatched bars, $n = 5–6$) for 15 min before stimulation with 300 μM NMDA. Data are expressed as mean % AUC ± S.E.M. * Significantly different than control, $P < 0.05$.

3.2. Effects of NO on basal and NMDA-stimulated [^{14}C]GABA and [^{3}H]ACh release

NMDA-stimulated neurotransmitter release has been shown to involve activation of nitric oxide synthase (Okhuma et al., 1995; Oh and McCaslin, 1996). To test the involvement of NO in NMDA-stimulated [^{14}C]GABA and [^{3}H]ACh release, striatal slices were superfused for 15 min with 10 μM nitroarginine (N-Arg), a nitric oxide synthase inhibitor, prior to stimulation with NMDA. The increase in basal [^{14}C]GABA release upon blocking the GABA transporter with 20 nM NNC-711 demonstrated in Fig. 1 was blocked by N-Arg by about 60%, thereby suggesting a role for NO in basal release of GABA in this preparation (Fig. 2). However, the modest increase in basal [^{3}H]ACh efflux caused by NNC-711 was not significantly affected by N-Arg (Fig. 2). It is possible that the increase in basal [^{3}H]ACh efflux caused by NNC-711 is due to increased extracellular GABA acting at $GABA_A$ receptors as we have observed that isoguvacine can potentiate basal [^{3}H]ACh efflux in a picrotoxin-sensitive manner (data not shown).

N-Arg also caused a significant 40% inhibition of NMDA-stimulated [^{14}C]GABA release, whereas [^{3}H]ACh release was only inhibited by 12% ($P > 0.05$) (Fig. 3A). Because guanylate cyclase often mediates the actions of NO, we utilized the selective inhibitor 1*H*-[1,2,4]oxadiazolo[4,4-a]quioxalin-1-one (ODQ) (Boulton et al., 1995; Garthwaite et al., 1995) to further investigate the role of the NO/guanylate cyclase/cGMP pathway in NMDA-stimulated transmitter release. After superfusing the slices with 1 μM ODQ for 15 min prior to NMDA challenge, NMDA-stimulated [^{14}C]GABA and [^{3}H]ACh release were blocked by 70% and 30%, respectively (Fig. 3B).

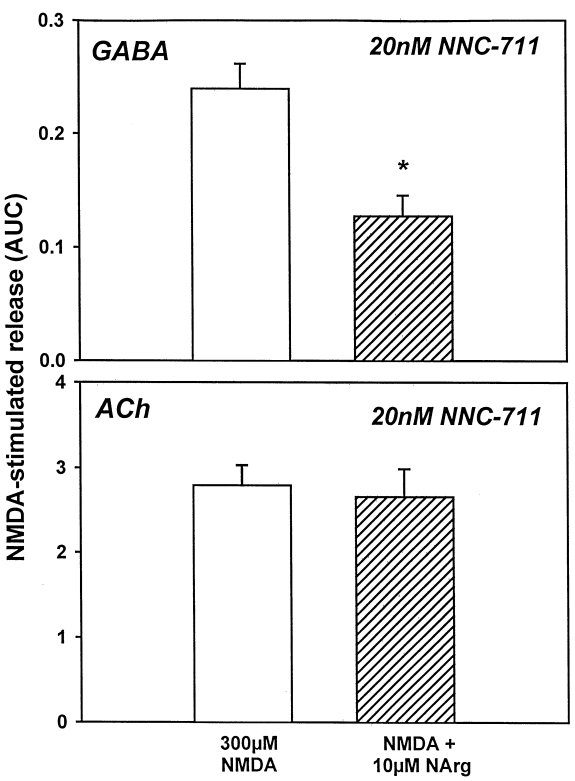

Fig. 4. The effect of nitroarginine (N-Arg) on of NMDA-stimulated [^{14}C]GABA and [^{3}H]ACh release in the presence of NNC-711. Striatal slices were superfused with 20 nM NNC-711 alone (open bars, $n = 8–12$) or with NNC-711 + 10 μM N-Arg (hatched bars, $n = 11–12$) 25 min prior to stimulation with 300 μM NMDA. The data are expressed as mean AUC ± S.E.M. * Significantly different than control, $P < 0.05$.

3.3. Effect of NNC-711 on NO-stimulated release

Previous reports from our laboratory showed that NO-stimulated catecholamine release was almost completely prevented by transport inhibitors, suggesting that reversal of the transporter was an important mechanism by which NO facilitated transmitter efflux (Lonart and Johnson, 1994, 1995b). In this study, NNC-711, a selective inhibitor of GABA transporter 1 (also known as GAT-1 and GAT-B), the predominant neuronal GABA transporter in the striatum (Clark et al., 1992), was used in a manner analogous to desipramine and nomifensine in our earlier investigations of NO-induced norepinephrine and dopamine release. At a concentration approximately four times its K_i, NNC-711 did not alter the degree to which NMDA-stimulated [^{3}H]GABA efflux was inhibited by N-Arg (Figs. 4 and 3A). This suggests that reversal of the neuronal GABA transporter in the striatum is not involved in the NO component of NMDA-evoked GABA efflux.

4. Discussion

Before discussing the major experimental results of this study, we would like to briefly discuss the results of our characterization of this system with NNC-711 and tetrodotoxin. In rat brain synaptosomes, NNC-711 has been shown to be a selective and potent GABA uptake inhibitor (Suzdak et al., 1992). It is also known to selectively inhibit the predominant neuronal GABA transporter in the striatum (Clark et al., 1992). Upon superfusion with NNC-711, we observed an increase in basal [^{14}C]GABA efflux which was blocked by the sodium channel blocker tetrodotoxin (Fig. 1). This suggests that there is an ongoing uptake of [^{14}C]GABA released by spontaneously active neurons in the slice preparation. In addition, it has been shown that stimulated GABA release can occur via classical exocytosis or via a calcium-independent mechanism, possibly involving reversal of the transporter (Bernath and Zigmond, 1988; Agostinho et al., 1994). Previous reports from this laboratory have shown that 80% of NMDA-

stimulated endogenous striatal GABA release was inhibited by 1 μM NNC-711 (Wang et al., 1996). In the present study it was found that almost 50% of NMDA-stimulated [^{14}C]GABA release is inhibited by 20 nM NNC-711, again suggesting that this release is partially mediated via reversal of the GABA transporter. Reports from other laboratories using K^+ or electrical field stimulation also support these findings; for example, nipecotic acid or NNC-711 has been shown to inhibit the calcium-independent component of electrically or K^+-stimulated [^{3}H]GABA or endogenous GABA release from striatal slices and synaptosomes (Bernath and Zigmond, 1988; Turner and Goldin, 1989; Agostinho et al., 1994; Wang et al., 1996). In this system a portion of basal efflux and almost all of the NMDA-stimulated [^{14}C]GABA efflux is tetrodotoxin-sensitive. Further, about one-half of the NMDA stimulated [^{14}C]GABA efflux is mediated by reversal of the neuronal transporter and is most likely not Ca^{2+}-dependent, though this is difficult to demonstrate because of the membrane destabilization caused by removal of Ca^{2+} in the absence of Mg^{2+}.

In rat striatum, a class of striatal interneurons contain nitric oxide synthase (Kawaguchi, 1993, 1997) and although NO is thought to be involved in neurotransmitter release in the striatum, the data supporting this notion is still controversial. There are several studies that have demonstrated that NO or NO donors increase striatal transmitter release, but the role of endogenous NO and/or cGMP is not yet well understood. Semba et al. (1995) demonstrated that inhibition of NOS actually increased the extracellular levels of striatal GABA as estimated by in vivo microdialysis, thereby suggesting an inhibitory role for NO. On the other hand, Segovia et al. (1994) found no effect of NOS inhibition on GABA efflux in a similarly designed experiment. In vitro experiments using striatal slices have also produced inconsistent results. That is, Moller et al. (1995) demonstrated that NMDA-induced [^{3}H]GABA efflux was enhanced by both the NOS substrate, L-arginine, and the competitive NOS inhibitor, NArg. These data are difficult to reconcile, but it is possible that the effect of L-arginine is unrelated to NO production as recently demonstrated for the effects of L-arginine on striatal DA efflux in vivo (Silva et al., 1998). NO donors might be expected to have the opposite effect of NOS inhibitors. However, Moller et al. (1995) observed that three different NO donors failed to affect NMDA-stimulated [^{3}H]GABA release. Thus, the role of NO in striatal GABA release is still unclear.

In this study, N-Arg significantly blocked 40% of NMDA-stimulated [^{14}C]GABA efflux without significantly altering NMDA-stimulated [^{3}H]ACh release. The guanylate cyclase (GC) inhibitor ODQ, however, blocked 70% of NMDA-stimulated [^{14}C]GABA and 30% of NMDA-stimulated [^{3}H]ACh release (Fig. 3). The greater effectiveness of ODQ could be due to an incomplete inhibition of NOS by superfusion with 10 μM N-Arg, though we, like others, have previously demonstrated that this concentration completely blocked NMDA-induced NOS activity or cGMP increases in a static incubation assay (Alagarsamy et al., 1994; East et al., 1996). In addition, inhibition of an effector molecule such as guanylate cyclase that is further downstream in an amplification cascade might be more effective than inhibition more upstream, particularly if the upstream effector molecule is not completely inhibited. These data suggest that in this system, NMDA-stimulated [^{14}C]GABA release and, to a lesser extent, [^{3}H]ACh release is partially mediated by NO. This agrees with experiments showing that L-arginine increases striatal [^{3}H]GABA efflux evoked by NMDA, but not with other experiments from the same laboratory that show that NMDA-stimulated [^{3}H]GABA release is potentiated by NOS and GC inhibitors (Moller et al., 1995). Those experiments, however, were conducted in the presence of 100 μM nipecotic acid. Since nipecotic acid is known to be transported by the GABA transporter (Johnston et al., 1976; Falch et al., 1990), basal GABA release could have substantially increased and the subsequent effect of NMDA stimulation altered in an unpredictable manner. Another potentially important, related difference between these experiments is that NMDA-stimulated GABA efflux in the presence of GABA transport inhibitors, including nipecotic acid, is significantly more Ca^{2+}-dependent than NMDA-induced release in the absence of transport inhibitors (Wang et al., 1996). Thus, it is possible that NO may differentially affect GABA released by classic Ca^{2+}-dependent exocytosis and by non-classic glutamate (or NMDA)-induced reversal of the GABA transport.

Previous reports from our laboratory demonstrated that NO donors such as hydroxylamine and nitrosocysteine (NO-CYS) evoked both [^{3}H]dopamine and [^{3}H]norepinephrine release from striatal and hippocampal slices in a manner that was blocked by monoamine uptake inhibitors such as desipramine, nomifensine and mazindol (Lonart and Johnson, 1994, 1995b). Furthermore, NO-CYS also inhibited both [^{3}H]dopamine and [^{3}H]norepinephrine uptake in striatal and hippocampal synaptosomes. Those data strongly suggested that NO donors cause norepinephrine release via reversal of the norepinephrine transporter (Lonart and Johnson, 1995b). On the other hand, the present data show that even in the presence of NNC-711, N-Arg inhibited NMDA-stimulated [^{14}C]GABA release to a similar extent as in its absence (Figs. 4 and 3A). This suggests that NO-mediated [^{14}C]GABA release does not occur via reversal of the GABA transporter. Interestingly, the increase in basal GABA efflux caused by blocking the GABA transporter was also blocked by about 50% by N-Arg (Fig. 2), thereby suggesting that NO is also involved in the ongoing basal release of GABA from striatal neurons.

These results highlight the differences between the stimulatory effects of the nitric oxide synthase/cGMP pathway on [^{14}C]GABA release observed here and the

apparent inhibitory effects of NO previously reported (Moller et al., 1995). That is, the inhibitory effect of N-Arg in the absence or presence of 20 nM NNC-711 appears to be inconsistent with the hypothesis that NO differentially affects classical and non-classical (transporter reversal) release. However, comparison of these experiments with 20 nM NNC-711 (a concentration 3.5 times its K_i) may not be directly comparable to nipecotic acid (at a concentration 15 times its K_i). Not only is nipecotic acid less selective for the major neuronal GABA transporter found in the striatum, it is also believed to be transported, while compounds structurally related to NNC-711 are not (Johnston et al., 1976; Falch et al., 1990; Suzdak et al., 1992). Although the mechanism is unknown, it seems possible that the use of nipecotic acid at a relatively high concentration could alter the dynamics of NMDA-induced GABA release in a manner that could account for the apparent inhibitory effect of NO observed by Moller et al. (1995).

In summary, this study has demonstrated that basal and NMDA-stimulated [^{14}C]GABA efflux involves a complex mechanism that includes reversal of the GABA transporter as well as activation of the NO/cGMP pathway. In contrast to studies showing a stimulatory effect of N-Arg on NMDA-stimulated GABA release, we showed that N-Arg and ODQ inhibit NMDA-stimulated GABA release, suggesting a stimulatory role for NO on striatal GABA release. In addition, this study provided evidence showing that the mechanisms underlying NO-mediated GABA release does not involve reversal of the GABA transporter, a mechanism that has been shown to be involved in the release of catecholamines. Finally, activation of nitric oxide synthase containing striatal interneurons by NMDA receptor stimulation appears to have a greater potential for influencing GABA release than ACh release.

Acknowledgements

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References

Agostinho, P., Duarte, C.B., Carvalho, A.P., Oliveira, C.R., 1994. Effect of oxidative stress on the release of [^{3}H]GABA in cultured chick retina cells. Brain Res. 655, 213–221.

Alagarsamy, S., Lonart, G., Johnson, K.M., 1994. Regulation of nitric oxide synthase activity in cortical slices by excitatory amino acids and calcium. J. Neurosci. Res. 38, 648–653.

Albin, R.L., 1995. The pathophysiology of Chorea/Ballism and Parkinsonism. Parkinsonism Relat. Disord. 1, 3–11.

Angulo, J.A., McEwen, B.S., 1994. Molecular aspects of neuropeptide regulation and function in the corpus striatum and nucleus accumbens. Brain Res. Rev. 19, 1–28.

Bernath, S., Zigmond, M.J., 1988. Characterization of [^{3}H]GABA release from striatal slices: evidence for a calcium-independent process via the GABA uptake system. Neuroscience 27, 563–570.

Bohme, G.A., Bon, C., Stulzmann, J.M., Doble, A., Blanchard, J.C., 1991. Possible involvement of nitric oxide in long-term potentiation. Eur. J. Pharmacol. 199, 379–381.

Boulton, C.L., Irving, A.J., Southam, E., Potier, B., Garthwaite, J., Collingridge, G.L., 1994. The nitric oxide-cyclic GMP pathway and synaptic depression in rat hippocampus slices. Eur. J. Neurosci. 6, 528–1535.

Boulton, C.L., Southam, E., Garthwaite, J., 1995. Nitric oxide-dependent long-term potentiation is blocked by a specific inhibitor of soluble guanylyl cyclase. Neuroscience 69, 703–966.

Bredt, D.S., Snyder, S.H., 1989. Nitric oxide mediates glutamate-linked enhancement of cGMP levels in the cerebellum. Proc. Natl. Acad. Sci. USA 86, 9030–9033.

Clark, J.A., Deutch, A.Y., Gallipol, P.F., Amara, S.G., 1992. Functional expression and CNS distribution of a β-alanine-sensitive neuronal transporter. Neuron 9, 337–348.

Di Chiara, G., Morelli, M., Consolo, S., 1994. Modulatory functions of neurotransmitters in the striatum: ACh/dopamine/NMDA interactions. Trends. Neurosci. 17, 228–233.

East, S.J., Parry-Jones, A., Brotchie, J.M., 1996. Ionotropic glutamate receptors and nitric oxide synthesis in the rat striatum. NeuroReport 8, 71–75.

Falch, E., Larsson, O.M., Schousboe, A., Krogsgaard-Larsen, P., 1990. $GABA_A$ agonists and GABA uptake inhibitors: structure-activity relationships. Drug Dev. Res. 21, 169–188.

Garthwaite, J., Charles, S.L., Chess-Williams, R., 1988. Endothelium derived relaxing factor release on activation of NMDA receptor suggests role as intercellular messenger in the brain. Nature 336, 385–388.

Garthwaite, J., Garthwaite, G., Palmer, R.M.J., Moncada, S., 1989. NMDA receptor activation induces nitric oxide synthesis from arginine in rat brain slices. Eur. J. Pharmacol. 172, 413–416.

Garthwaite, J., Southam, E., Boulton, C.L., Nielsen, E.B., Schmidt, K., Mayer, B., 1995. Potent and selective inhibition of nitric oxide-sensitive guanylyl cyclase by 1*H*-[1,2,4]Oxadiazolo[4,4-a]quioxalin-1-one. Mol. Pharmacol. 48, 184–188.

Graybiel, A.M., 1990. Neurotransmitters and neuromodulators in the basal ganglia. Trends Neurosci. 13, 244–254.

Guevara-Guzman, R., Emson, P.C., Kendrick, K.M., 1994. Modulation of in vivo striatal transmitter release by nitric oxide and cyclic GMP. J. Neurochem. 62, 807–810.

Hammond, E.J., Wilder, B.J., 1985. Gamma-vinyl GABA. Gen. Pharmacol. 16, 441–447.

Hirsch, D.B., Steiner, J.P., Dawson, T.M., Mammen, A., Hayek, E., Snyder, S.H., 1993. Neurotransmitter release regulated by nitric oxide in PC-12 cells and brain synaptosomes. Curr. Biol. 3, 749–752.

Johnston, G.A.R., Krogsgaard-Larsen, P., Stephanson, A.L., Twitchin, B., 1976. Inhibition of the uptake of GABA and related amino acids in rat brain slices by the optical isomers of nipecotic acid. J. Neurochem. 26, 1029–1032.

Kawaguchi, Y., 1993. Physiological, morphological and histochemical characterization of three classes of interneurons in rat neostriatum. J. Neurosci. 13, 4908–4923.

Kawaguchi, Y., 1997. Neostriatal cell subtypes and their functional roles. Neurosci. Res. 27, 1–8.

Landwehrmeyer, G.B., Standaert, D.G., Testa, C.M., Penny Jr., J.B., Young, A.B., 1995. NMDA receptor subunit mRNA expression by projection neurons and interneurons in rat striatum. J. Neurosci. 15, 5297–5307.

Lonart, G., Johnson, K.M., 1994. Inhibitory effects of nitric oxide on the uptake of [^{3}H]dopamine and [^{3}H]glutamate by striatal synaptosomes. J. Neurochem. 63, 2108–2117.

Lonart, G., Johnson, K.M., 1995a. Characterization of nitric oxide generator-induced hippocampal [^{3}H]norepinephrine release: I. The role of glutamate. J. Pharmacol. Exp. Ther. 275, 7–13.

Lonart, G., Johnson, K.M., 1995b. Characterization of nitric oxide generator-induced hippocampal [^{3}H]norepinephrine release: II. The role of calcium, reverse norepinephrine transport and cyclic 3′5′-guanosine monophosphate. J. Pharmacol. Exp. Ther. 275, 14–22.

Lonart, G., Wang, J., Johnson, K.M., 1992. Nitric oxide induces neurotransmitter release from hippocampal slices. Eur. J. Pharmacol. 220, 271–272.

Lonart, G., Cassels, K.L., Johnson, K.M., 1993. Nitric oxide induces calcium-dependent [^{3}H]dopamine release from striatal slices. J. Neurosci. Res. 35, 192–198.

Moller, M., Jones, N.M., Beart, P.M., 1995. Complex involvement of nitric oxide and cGMP at *N*-methyl-D-aspartic acid receptors regulating γ-[^{3}H]aminobutyric acid release from striatal slices. Neurosci. Lett. 190, 195–198.

Montague, P.R., Gancayco, C.D., Winn, M.J., Marchase, R.B., Friedlander, M.J., 1994. Role of NO production in NMDA receptor-mediated neurotransmitter release in cerebral cortex. Science 263, 973–977.

O'Dell, T.J., Hawkins, R.D., Kandel, E.R., Arancio, O., 1991. Tests of the roles of two diffusible substances in long-term potentiation: evidence for nitric oxide as a possible early retrograde messenger. Proc. Natl. Acad. Sci. USA 88, 11285–11289.

Oh, S., McCaslin, P.P., 1996. Nitric oxide synthase inhibitor decreases the *N*-methyl-D- aspartate-induced release of glutamate in cerebellar granule neurons. Med. Sci. Res. 24, 69–71.

Okhuma, S., Katsura, M., Chen, D., Narihara, H., Kuriyama, K., 1995. Facilitation of *N*-methyl-D-aspartate-evoked acetylcholine release by hydroxyl scavengers. NeuroReport 6, 2033–2036.

Penney, J.B., Standaert, D.G., Testa, C.M., Landwehrmeyer, G.B., Young, A.B., 1996. Glutamate receptor genes in Parkinson's disease. In: Battistin, L., Scarlato, G., Caraceni, T., Ruggieri, S. (Eds.), Advances in Neurology. Lippincott-Raven Press, Philadelphia, PA, pp. 79–86.

Prast, H., Philippu, A., 1992. Nitric oxide releases acetylcholine in the basal forebrain. Eur. J. Pharmacol. 216, 139–140.

Sandor, N.T., Brassai, A., Puskas, A., Lendvai, B., 1995. Role of nitric oxide in modulating neurotransmitter release from rat striatum. Brain Res. Bull. 36, 483–486.

Schuman, E.M., Madison, D.V., 1991. A requirement for the intercellular messenger nitric oxide in long-term potentiation. Science 254, 1503–1506.

Segovia, G., Porras, A., Mora, F., 1994. Effects of a nitric oxide donor on glutamate and GABA release in striatum and hippocampus of the conscious rat. NeuroReport 5, 1937–1940.

Semba, J., Sakai, M., Miyoshi, R., Kito, S., 1995. N^G-monomethyl-L-arginine, an inhibitor of nitric oxide synthase, increases extracellular GABA in the striatum of the freely moving rat. NeuroReport 6, 1298–1300.

Shibuki, K., Okada, D., 1991. Endogenous nitric oxide release required for long-term synaptic depression in the cerebellum. Nature 349, 326–328.

Silva, M.T., Rose, S., Hindmarsh, J.G., Jenner, P., Marsden, C.D., 1998. L-arginine produces NO-independent increases in dopamine efflux in rat striatum. NeuroReport 9, 149–152.

Southam, E., East, S.J., Garthwaite, J., 1991. Excitatory amino acid receptors coupled to the nitric oxide/cGMP pathway in rat cerebellum during development. J. Neurochem. 56, 2072–2081.

Stout, A.K., Woodward, J.J., 1994. Differential effects of nitric oxide gas and nitric oxide donors on depolarization-induced release of [^{3}H]norepinephrine from rat hippocampal slices. Neuropharmacology 33, 1367–1374.

Suzdak, P.D., Frederiksen, K., Andersen, K.E., Sorensen, P.O., Knutsen, L.J.S., Nielsen, E.B., 1992. NNC-711, a novel potent and selective γ-aminobutyric acid uptake inhibitor: pharmacological characterization. Eur. J. Pharmacol. 223, 189–198.

Takita, M., Kaneko, H., Suzuki, S.D., Akamatsu, M., 1997. Lasting effect of NO on glutamate release in rat striatum revealed by continuous brain dialysis. NeuroReport 8, 567–570.

Turner, T.J., Goldin, S.M., 1989. Multiple components of synaptosomal [^{3}H] γ-aminobutyric acid release resolved by a rapid superfusion system. Biochemistry 28, 586–593.

Wang, J., Lonart, G., Johnson, K.M., 1996. Glutamate receptor activation induces carrier mediated release of endogenous GABA from rat striatal slices. J. Neural Transm. 103, 31–43.