NeuroImage 142 (2016) 663–667

Contents lists available at ScienceDirect

NeuroImage

journal homepage: www.elsevier.com/locate/ynimg

Comments and Controversies

Prefrontal cortex dopamine release measured in vivo with positron emission tomography: Implications for the stimulant paradigm

D. Hernaus[a,*], M.A. Mehta[b]

[a] Department of Psychiatry and Neuropsychology, South Limburg Mental Health Research and Teaching Network, EURON, Maastricht University Medical Centre, Maastricht, The Netherlands
[b] Centre for Neuroimaging Sciences, Institute of Psychiatry, Psychology & Neuroscience, King's College, London, United Kingdom

ARTICLE INFO

Article history:
Accepted 18 June 2015
Available online 26 June 2015

Keywords:
Dopamine
PET
Amphetamine
Stimulants
$D_{2/3}$
Striatum
PFC

ABSTRACT

Experimental tasks and stimulant paradigms in combination with $D_{2/3}$ emission tomography have been essential in understanding the dopamine (DA) system. However, whereas task-induced DA release is dependent on a mechanism that is largely similar throughout the brain, the DA-increasing stimulant mechanism of action changes drastically from striatum to cortex. We posit the problems that may be encountered when translating the stimulant emission tomography paradigm from striatum to PFC. After comparing the available human data on task- and stimulant-induced changes in extracellular PFC DA assessed with PET, we hypothesize that the stimulant paradigm in the PFC, even with high affinity tracers, may not completely capture the true effect of stimulants on extracellular PFC DA levels. Task-induced and stimulant-induced effects on extracellular PFC DA measured with emission tomography should therefore be regarded as different phenomena. We conclude with future directions and alternative probes to measure PFC DA transmission with emission tomography.

Introduction

Investigation of the dopamine (DA) system in vivo is critical to understanding its role in health and disease. The focus on cortical, particularly prefrontal cortex (PFC), DA has revealed an essential role in working memory (Arnsten et al., 1994), clinical response to stimulants (Arnsten, 2006), and memory-guided action (Wang et al., 2004). While the importance of PFC DA transmission in many psychological functions is acknowledged, in vivo investigation of the DA system with molecular imaging techniques (PET and SPECT) has for the most part remained restricted to areas with high DA receptor density, such as the striatum.

The established method to assess the functional properties of the striatum in vivo utilises molecular imaging combined with a DA receptor subtype 2 and 3 ($D_{2/3}$) sensitive tracer and task (Egerton et al., 2009) or stimulant paradigm (Laruelle, 2000). It is presumed that task-induced DA release is governed by endogenous mechanisms: this is demonstrated by the observation that tasks affect DA synthesis (Demarest et al., 1985; Freed and Yamamoto, 1985; Hattori et al., 1994) and DA release into the synaptic cleft (Cousins and Salamone, 1996; Finlay et al., 1995; McCullough and Salamone, 1992), corresponding to *increased* cell firing (Dugast et al., 1994; Garris et al., 1994). In contrast, stimulants increase extracellular DA via a range of direct and indirect mechanisms. First, the extracellular DA-enhancing effects of stimulants are mediated via DA, norepinephrine (NE) and serotonin transporter (DAT; NET; SERT) blockade; this has been demonstrated in striatum and PFC (Bymaster et al., 2002; Jones et al., 1999). Second, stimulants also increase extracellular DA via transporter reversal (Robertson et al., 2009; Sulzer et al., 1993; Giambalvo, 2003), internalisation (Boudanova et al., 2008; Melikian and Buckley, 1999) and trace-amine associated receptor 1 (TAAR1) agonism (Miller, 2011; Reese et al., 2007), primarily validated in the basal ganglia. It is noteworthy, however, that stimulants *decrease* overall DA cell activity via regulatory feedback (Bunney et al., 1973; Shi et al., 2000). Thus, whereas tasks and stimulants are both used to investigate brain DA function, they act via markedly different ways to promote DA release.

Stimulant (Laruelle, 2000; Volkow et al., 2002a) and task (Egerton et al., 2009) molecular imaging paradigms have been successfully employed in the striatum and, recently, have also been used to explore the functional properties of PFC DA release in vivo. However, whereas the endogenous mechanism by which tasks increase extracellular DA levels remains similar throughout the brain (Freed and Yamamoto, 1985; Hattori et al., 1994; Finlay et al., 1995; Abercrombie et al., 1989), stimulant mechanisms of action differ substantially between striatum and PFC (Stahl, 2003). This may affect the suitability of the stimulant paradigm to investigate DA function in extra-striatal areas. An important question therefore is: how valid is it to translate the stimulant paradigm to the PFC? In this commentary, we aim to address the differences in stimulant mechanisms of action between striatum and PFC and its implications for the stimulant molecular imaging paradigm in the PFC.

A general challenge that exists for molecular imaging paradigms (task *and* stimulant) is the low density of $D_{2/3}$ in the PFC, compared to the striatum (Hall et al., 1994). This is because receptor density is an

* Corresponding author.
E-mail addresses: dennis.hernaus@maastrichtuniversity.nl (D. Hernaus), Mitul.Mehta@kcl.ac.uk (M.A. Mehta).

http://dx.doi.org/10.1016/j.neuroimage.2015.06.057
1053-8119/

essential factor in determining the signal-to-noise ratio of emission tomography data (Hall et al., 1994; Farde et al., 1988). The low signal-to-noise ratio for a typical D_2 tracer such as (McCullough and Salamone, 1992) [11] C-raclopride in the cortex (Hall et al., 1988) can be overcome by using higher-affinity tracers (Halldin et al., 1995). Still, even with higher affinity tracers, the complex mechanisms of actions of stimulants may differentially impact measurements in the PFC in drug challenge relative to task studies.

One key mechanism by which stimulants increase extracellular striatal DA is the DAT (Bymaster et al., 2002; Moron et al., 2002). However, DAT expression in the PFC is lower than in the striatum (Richtand et al., 1995; Sesack et al., 1998) and, subsequently, less efficient in regulating extracellular DA levels (Moron et al., 2002). In the cortex, the NET *also* regulates stimulant-induced increases in extracellular DA. This is demonstrated by the observations that AHN 2-005 (Schmeichel et al., 2013) (DAT-inhibitor), atomoxetine (Bymaster et al., 2002) (NET-inhibitor), methylphenidate (Bymaster et al., 2002) (DAT- and NET-inhibitor), but not fluoxetine (Bymaster et al., 2002; Shi et al., 2000) (SERT-inhibitor), increase extracellular PFC DA to a similar extent (>200%). Comparison studies even suggest that NET-blockade may be more efficient in increasing PFC DA levels than DAT-blockade (Carboni et al., 2006; Masana et al., 2011).

Importantly, these studies all utilized microdialysis, a technique that samples neurotransmitters far from the synaptic cleft (Chefer et al., 2006). This is noteworthy, because the NET is *not* localised on DA neurons (Schroeter et al., 2000). Moreover, NA neurons are also responsible for DA release in the cortex: this has been observed in PFC (Devoto et al., 2001) and hippocampus (Smith and Greene, 2012). Lastly, PFC DAT are expressed in pre-terminal regions rather than peri-synaptically (Sesack et al., 1998). This all suggests that a substantial proportion of the stimulant-induced increases in extracellular PFC DA occur outside of the DA synapse. PFC DA release measurement using emission tomography thus becomes at least partially dependent on *diffusion* (Sesack et al., 1998; Cragg et al., 2001) to displace the $D_{2/3}$-bound tracer, which to a significant degree is located in the synapse (Laruelle, 2000). Moreover, these mechanisms of action may render stimulant-induced increases in extracellular PFC DA particularly susceptible to the catechol-O-methyltransferase (COMT) enzyme, an important regulator of extracellular DA in the cortex (Mannisto and Kaakkola, 1999).

Another mechanism by which stimulants increase extracellular PFC DA is related to ventral tegmental area (VTA) cell firing: administration of amphetamine into the VTA dose-dependently increases extracellular PFC DA levels (Pan et al., 1996). Moreover, NET inhibition increases VTA cell burst firing (Shi et al., 2000; Linner et al., 2001), which has been co-observed with increased extracellular PFC DA levels (Linner et al., 2001). Three important observations, however, need to be considered here. First, amphetamine decreases overall DA cell firing, most likely via D_2 autoreceptors (Bunney et al., 1973; Shi et al., 2000). Second, TAAR1 agonism, another important mechanism of stimulants, *also* decreases VTA cell firing (Revel et al., 2011). Third, within normal dose range, amphetamine decreases *locus coeruleus* (LC) cell firing, most likely via $\alpha 2$ autoreceptors (Curet et al., 1992; Engberg and Svensson, 1979; Huang and Maas, 1981). This last observation is especially noteworthy because $\alpha 2$ autoreceptor blockade of LC NA cells reverses increased PFC DA levels after intra-VTA administration of amphetamine (Pan et al., 1996). Thus, whereas some stimulant mechanisms of action may promote PFC DA release when applied regionally or in isolation, there are multiple mechanisms that inhibit VTA cell firing. VTA DA cell firing may increase PFC DA levels to a certain extent, but it is unlikely to be sufficient to explain the amphetamine-induced increase in PFC DA levels.

A final difference between stimulant mechanisms of action in striatum and PFC is timing-related. Whereas amphetamine produces a sharp increase in extracellular caudate DA in non-human primates, amphetamine-induced increases in extracellular PFC DA are gradual and prolonged, again, suggestive of regional differences in regulatory mechanisms (Jedema et al., 2014).

Thus, it seems that stimulant-induced increases in extracellular PFC DA, unlike the striatum, depend on significant *diffusion, complex mechanisms not always in the vicinity of the $D_{2/3}$ and are characterized by a slower, more gradual, time profile.* This contrasts with task-induced DA release, which is governed by similar endogenous mechanisms in striatum and PFC and makes use of a relatively direct, or synaptic, route (Egerton et al., 2009). Still, with high affinity ligands such as FLB 457, amphetamine-induced changes in extracellular PFC DA can be detected (Narendran et al., 2014a). In the non-human primate, this corresponds to the magnitude of amphetamine-induced increases in extracellular PFC DA sampled with microdialysis (Narendran et al., 2014a). However, two important questions that remain are:

- Are the quantities of stimulant-induced increases in extracellular PFC DA measured with PET an accurate reflection of the true effect of stimulants on PFC DA in the extracellular space and/or the terminal fields of DA neurons?
- Are the mechanisms through which stimulants increase extracellular PFC DA adequate to index with PET?

These questions can be answered empirically using all studies to date in healthy volunteers that have used a placebo-stimulant/control-experimental within-subject design with FLB 457 and fallypride (Fig. 1). Given the complex mechanism of stimulants, but not tasks, our prediction is that stimulant-induced PFC DA release measured with PET is captured less uniformly and consistently than task-induced PFC DA release measured with PET.

Indeed, in the preliminary literature that is currently available, stimulant-induced PFC DA release measured with PET displays significant variation (Fig. 1). Stimulant-induced PFC DA release is consistently detected with the high-affinity ligand FLB 457, but less so with fallypride. The only human study available that directly compared fallypride to FLB 457 in the same participants confirmed the reduced sensitivity of fallypride to stimulant-induced changes in extracellular PFC DA (Narendran et al., 2009). However, stimulant-induced DA release measured with fallypride has been reported in greater sample sizes (Zald and Treadway, 2015) and in regions that show the highest cortical DAergic innervation (Zald and Treadway, 2015; Cropley et al., 2008), such as the anterior cingulate (Camus et al., 1986; Tassin et al., 1978) and medial frontal cortex (Berger et al., 1976; Emson and Koob, 1978; Vincent et al., 1993) (Fig. 1).

In the absence of empirical in vivo data and using the only data available, task-induced tracer displacement in PFC (working memory, vigilance (Aalto et al., 2005), response inhibition (Anstrom and Woodward, 2005) and stress (Nagano-Saito et al., 2013)) is similar for fallypride and FLB 457 (Fig. 1). Interestingly, task-induced displacement values are similar or greater than those observed with stimulants and, for fallypride, are not influenced by blood flow (Ceccarini et al., 2012; Cumming et al., 2013). Thus, the relatively direct route of task-based DA release may suggest that fallypride and FLB 457 could have similar utility in the PFC for task paradigms. A direct comparison of the sensitivity of these two ligands for measuring task-induced changes in extracellular DA and how this relates to actual extracellular PFC DA levels is urgently needed. Although these data suggest that task-induced PFC DA release measured with PET is more uniformly and consistently captured than stimulant-induced PFC DA release, it should be noted that tracer affinity (FLB 457 > fallypride) may also explain some of these results.

The discrepancy between stimulant- and task-induced DA release measured with PET may be related to regional variation in stimulant versus task mechanisms of action. Blockade of abundant DAT (Volkow et al., 2002a, 2002b) and the exceptionally high DA innervation in the striatum (Cortes et al., 1989) may ensure that stimulant-induced increases in extracellular DA are uniformly captured with PET here. The complex route in the cortex, however, may promote stimulant-induced DA release outside of the vicinity of the $D_{2/3}$, negatively affecting