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Pharmacokinetic Limitations on Effects of an Alpha7-Nicotinic Receptor Agonist in Schizophrenia: Randomized Trial with an Extended-Release Formulation

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The aim of the trial was to assess whether extending plasma levels of the alpha7-nicotinic acetylcholine receptor (nAChR) agonist 3-(2,4-dimethoxybenzylidene)-anabaseine (DMXB-A) over time enhances its cognitive effects in schizophrenia. Both smoking and non-smoking patients were studied, to determine whether effects differ between these two groups. Forty-three smokers and thirty-seven non-smokers who met DSM-IV criteria for schizophrenia were enrolled in a double-blind, randomized, placebo-controlled 1 month trial. DMXB-A 150 mg was formulated with hypromellose to produce extended release over 4 h and administered four times daily. The primary outcome (the Neurocognitive Composite of the MATRICS Consensus Cognitive Battery) and secondary outcomes (the MATRICS Attention-Vigilance Domain and P50 gating), showed no significant effect. Plasma levels were obtained 2.5 h post administration. In non-smokers, levels were similar to those reached transiently with 75–150 mg DMXB-A immediate-release formulations twice daily, which were earlier shown to be effective doses. However, the extended-release formulation produced no cognitive or clinical effect either in non-smokers or smokers. The 10-fold lower DMXB-A plasma levels in smokers suggest that chronic smoking enhances DMXB-A metabolism. Pro-cognitive effects of DMXB-A may result from transient increases in cell signaling that are limited by receptor tachyphylaxis. Future efforts to improve cognition in schizophrenia by enhancing alpha7 nAChR function may require consideration of these pharmacokinetic limitations.
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INTRODUCTION

The alpha7-nicotinic acetylcholine receptor (nAChR) has received increasing interest as a target for the development of new drugs that will ameliorate cognitive deficits in people with schizophrenia. Several companies developed drug candidates, some with successful initial trials (Lieberman *et al*, 2013; Keefe *et al*, 2015; Haig *et al*, 2016a). However, none of these candidates has shown significant cognitive effects in schizophrenia in larger trials (Umbricht *et al*, 2014; Walling *et al*, 2016; Haig *et al*, 2016b). We earlier showed cognitive effects with single-dose administration of the partial agonist 3-(2,4-dimethoxybenzylidene)-anabaseine (DMXB-A) using the Repeatable Battery for the Assessment of Neurocognitive Status, and after 1 month administration in the Attention and Executive Function Domains of the MATRICS Consensus Cognitive Battery (Olincy *et al*, 2006; Freedman *et al*, 2008). Unlike the relatively long plasma half-life drug candidates in industry development that were designed to permit daily dosing, DMXB-A has an exceptionally short half-life, ~60 min (Kitigawa *et al*, 2003). A short half-life would seem to be an undesirable property for a drug developed to treat a chronic illness. However, the alpha7-nAChR displays very rapid desensitization with even brief exposure to agonists (Peng *et al*, 1994; Lopez-Hernandez *et al*, 2009). Therefore, a short half-life drug that can produce a large transient stimulation of the receptor might actually produce a greater pro-cognitive effect relative to a drug producing a more constant elevation in receptor stimulation.

Differences in effect between drugs depend on both their pharmacodynamic and pharmacokinetic (PK) properties. Alpha7-nicotinic agonists differ not only in their half-lives but also in their relative effectiveness as agonists and antagonists, as all nAChR agonists, including acetylcholine itself, have both properties (Briggs *et al*, 1998; Papke *et al*, 2009). Therefore, comparison between drugs with different pharmacodynamic properties cannot answer the question of which PK properties are most desirable. This question has

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importance for developing drugs with optimal effectiveness. To determine which PK properties are optimal for an alpha7 agonist, we have tested an extended-release formulation of DMXB-A, the same partial agonist that we initially had tested as an immediate-release formulation.

DMXB-A was previously tested in a methylcellulose-filled gelatin capsule formulation that allows its immediate release in the stomach and maximal absorption in about 60 min (Kitagawa *et al*, 2003). To extend patient exposure to the compound, we reformulated it in hypromellose-filled capsules. Hypomellose swells into a matrix in the gastric fluid. DMXB-A is then released more slowly as the matrix is physically degraded during digestion. The process occurs more quickly in the alkaline duodenum than in the stomach, and the drug is released over 4 h in most patients. This extended-release formulation enables us to determine which formulation is optimal for DMXB-A, without the confound of differences in pharmacodynamic properties.

Heretofore, DMXB-A has been tested only in non-smoking patients, because of the potential for interference from the desensitization produced by nicotine and the possibility of increased adverse effects resulting from the simultaneous presence of both nAChR agonists. Industry trials have included smokers, but they have not assessed smoking itself to see whether the introduction of an agonist also changed smoking behavior, which indirectly might contribute to the cognitive effects. We therefore recruited half the subjects to be smokers to assess the effect of the slower release formulation on both their cognition and their smoking, measured as their nicotine and cotinine plasma levels at the conclusion of the 4-week treatment.

MATERIALS AND METHODS

Subjects

One hundred thirty-eight patients were screened. The most common exclusion was marijuana abuse. Eighty patients age 18–60 years with schizophrenia by DSM-IV criteria and SCID interview were enrolled, 37 non-smokers and 43 smokers (Table 1; Supplementary Figure S1). All subjects gave informed consent for the study.

Patients were currently being treated with a wide range of antipsychotic drugs in usual therapeutic doses, except for clozapine; treatment with clozapine was an exclusion criterion because it often alters inhibition of the P50-evoked potential, a biomarker for attention (Nagamoto *et al*, 1996). Two participants were not taking antipsychotic medication. All had been on the same antipsychotic regimen for at least 3 months without change in their clinical status. Their drug regimens were maintained during the trial. No patient required a change for clinical reasons. Exclusion criteria were history of severe head injury with neurocognitive disturbance, and positive urinary toxicology screen for drugs of abuse including marijuana. Female patients had an assured method of birth control and a negative pregnancy screen.

Patients were sub-grouped into smokers who smoked at least 10 cigarettes per day and non-smokers, who had not smoked for at least 1 month. Non-smoking status was verified by expired CO level.

Table 1 Demographic Data

	DMXB-A mean or *N*	SD	Placebo mean or *N*	SD
Age	46.5	12.4	43.2	13.2
Male	32		28	
Education	12.4	2.4	13.6	2.1
Caucasian (*N*)	31		27	
African-American (*N*)	6		10	
Hispanic (*N*)	1		3	
Other (*N*)	2		0	
Schizoaffective (*N*)	7		12	
Smokers (*N*)	21		22	
Packs per day	1.1	0.6	1.1	0.5
Total subjects (*N*)	40		40	

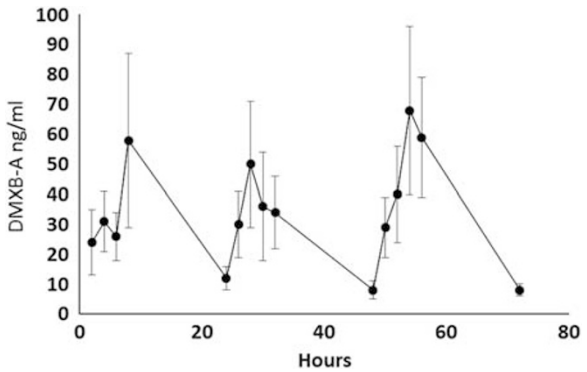

Figure 1 Mean and SD of 3-(2,4-dimethoxybenzylidene)-anabaseine (DMXB-A) plasma level during administration of 150 mg of the hypromellose formulation (DMXBA-ER) four times daily. There were $N = 10$ normal, non-smoking subjects who were not in the clinical trial.

Drug Formulation

3-(2,4-dimethoxybenzylidene)-anabaseine (DMXB-A) was synthesized as previously described (Olincy *et al*, 2006). For the extended-release preparation (DMXBA-ER), DMXBA 150 mg was mixed with hypromellose to fill a standard #1 capsule. Hypromellose is also used to formulate quetiapine for extended release. Automated filling and sealing techniques were employed. A preliminary study in 10 healthy non-smoking subjects who were not subjects in the subsequent clinical trial showed the time curve for change in the plasma concentration of DMXB-A. The mean level slowly increased between 1 and 4 h after administration (Figure 1). After administration four times daily, as was used in the current trial, no subject had levels before the next morning's dose >5 ng/ml. Thus, most DMXB-A is cleared from the plasma overnight.

Conduct of the Trial

The trial was approved by the Colorado Multi-Institutional Review Board. The trial was a parallel comparison between placebo and 150 mg DMXB-A-ER in equal numbers of subjects. All treatments and assessments were performed

under double-blind conditions at the University of Colorado from August 1, 2011 through June 1, 2015. Side effects and vital signs were monitored weekly, along with pill counts. All subjects received open-label placebo for 1 week. Subjects who could not maintain 70% compliance during that week were required to repeat the week to obtain compliance. Medication boxes were monitored weekly during the trial and compliance was encouraged. The MATRICS Consensus Cognitive Battery (MATRICS CCB), the primary outcome, and the secondary outcomes, BPRS, SANS (Sum of Global Ratings in Associality/Anhedonia, Affective Flattening, Avolition/Apathy, Alogia), and P50 auditory-evoked potential inhibition, were assessed at 4 weeks. Previously published methodology was used for these assessments (Olincy *et al*, 2006; Freedman *et al*, 2008; Nuechterlein *et al*, 2008). The planned enrollment was completed.

Plasma Drug Assays

Plasma specimens for drug level assays were obtained at 2 h after the first or second morning dose, after neurocognitive testing. The specimens were analyzed by high-performance liquid chromatography as previously described (Mahnir *et al*, 1998). DMXB-A's 4-hydroxy metabolite was generally below the level of reliable quantification.

Randomization and Statistical Analysis

Separate randomization schemes were used for smokers and non-smokers, to ensure equal numbers of drug and placebo treatments within each subgroup. The planned trial enrollment of 20 subject per group (DMXBA-ER or placebo, smokers or non-smoker) was determined by a power analysis (alpha 2 tailed 0.05, 1-beta = 0.8 based on the previous determination of DMXB-A's effect size ($d' = 0.97$) on the mean of MATRCS-CCB Neurocognitive domains (Freedman *et al*, 2008). Randomization was performed by the statistician using SAS and given to the pharmacists, who were the only non-blinded study personnel. The allocation sequence was not available to other personnel. The primary outcome measure was the MATRICS CCB Neurocognitive Composite, which encompasses all domains except Social Cognition (MATRICS Assessment Inc., 2015). An analysis of variance with sex, age, and baseline performance as covariates was performed for MATRICS CCB Neurocognitive Composite T-scores for assessment after 4 weeks of treatment. Protected univariate analyses were performed for each subgroup of smokers and non-smokers. All other exploratory analyses were performed with Student's *t*-tests. Reported significance levels are two-tailed.

Comparison with an Immediate-Release Formulation

An exploratory post hoc analysis to compare the effects of extended exposure (DMXBA-ER) with the previously studied immediate-release formulation (DMXBA-IR, Freedman *et al*, 2008) was conducted. The MATRICS CCB Neurocognitive Composite Index was not available at the time of the publication of that study. That Composite Index has now been computed. The earlier study had a three-arm crossover design with all subjects receiving DMXBA-IR 75 mg, 150 mg, and placebo. Data were previously analyzed from the first arm of the crossover compared with the baseline because of significant effects of repeated MATRICS CCB testing over time. For the *post hoc* analysis, the 16 patients who received DMXBA-IR in the first arm at either dose were combined. The mean DMXB-A plasma levels were skewed in both the present extended-release study and in the earlier immediate-release study, with one subject in each formulation group having plasma levels over three SDs above the mean. In the DMXBA-IR study, the subject had plasma levels three SDs above the mean for both doses. We hypothesize that these individuals failed to *O*-demethylate one or both of the DMXB-A methoxy groups, conversions that promote both agonist effects and excretion (Azuma *et al*, 1999; Kem *et al*, 2004). Therefore, the comparative analyses were conducted omitting these two individuals. None of the patients studied with the immediate-release formulation was smokers. For the extended-release formulation, only the non-smoking group was analyzed for the comparison.

RESULTS

Eighty patients with schizophrenia were studied for 4 weeks: 40 received extended-release DMXB-A 150 mg qid for 4 weeks and 40 received placebo. Sub-grouped by smoking status, 18 non-smokers received placebo and 19 received extended-release DMXB-A-ER; 21 smokers received placebo and 22 received DMXB-A-ER. One patient left the DMXBA-ER group because he had a transient ischemic episode. During evaluation in the Emergency Department, he revealed a history of several such episodes with similar clinical appearance, which he had not previously disclosed. His attack was not ascribed to his experimental treatment. A second patient withdrew after randomization and did not return for further evaluation. Reports of other adverse effects showed no significant differences between groups. The most common side effects in non-smokers who received DMXB-A-ER were gastrointestinal, including nausea, diarrhea, flatulence, and constipation. None was severe. These effects were not observed in smokers who received DMXB-A-ER. There was also a similar distribution of laboratory values outside the normal range for both groups. None was considered to be medically significant (Supplementary Tables 1). Neither DMXB-A-ER nor placebo groups displayed significant changes with weight, 0.8 ± 6.0 lbs for DMXB-A-ER and -0.3 ± 6.0 lbs for placebo.

Neurocognitive and Symptomatic Outcome

The primary outcome was the MATRICS CCB Neurocognitive Composite T score. An analysis of variance with age, sex, and pre-treatment baseline as covariates showed no significant relationship with any of these variables. The mean change from baseline for the non-smoker and smoker groups combined was 2.10 ± 8.24 for placebo and 0.64 ± 5.82 for DMXB-A-ER. The practice effect in the placebo group was non-significant ($t = 0.73$, df 39, $p = 0.47$). Subgroup analysis showed no effect in the non-smoking patients (placebo 2.78 ± 5.98 and DMXB-A-ER 1.06 ± 6.26) and no effect in the smoking patients (placebo 1.52 ± 9.90 and DMXB-A-ER 0.29 ± 5.43). No significant effects of treatment were observed on any MATRICS CCB individual domain.

Clinical ratings did not change significantly after DMXB-A-ER treatment: mean change in BPRS DXMBA-ER -3.11 ± 8.07, placebo -2.17 ± 4.63; mean change in SANS DMXBA-ER 0.22 ± 1.56, placebo 0.71 ± 1.81.

Inhibition of the P50 auditory-evoked potential was measured to assess whether DMXB-A-ER had its intended neurobiological effect. In the previous DMXB-A-IR trial, this had shown significant increase in inhibition with DMXBA-IR compared with placebo (Olincy *et al*, 2006). In the current trial, neither the smokers nor the non-smokers showed significant effects of DMXBA-ER on P50 inhibition, compared with placebo: DMXBA-ER P50 ratio 0.50 ± 0.56 compared with the placebo 0.39 ± 0.32.

There were no statistically significant differences in nicotine and cotinine levels in smokers with treatment. Nicotine levels were 14 ± 13 ng/ml during placebo treatment and 19.8 ± 16.1 ng/ml during DMXBA-ER. Cotinine levels were 122 ± 39 ng/ml during placebo treatment and 113.7 ± 39.3 ng/ml during DMXBA-ER.

Plasma levels of DMXB-A were an order of magnitude lower in smokers than in non-smokers. The levels were drawn at the time of cognitive testing, about 2.5 h after the morning dose. For non-smokers, the mean level was 46.7 ± 62.3 ng/ml and for smokers it was 4.2 ± 3.7 ng/ml.

Exploratory Comparison with the Immediate-Release Formulation

There was no significant difference in DMXB-A plasma levels between DMXB-ER (56.2 ± 96 ng/ml) and DMXBA-IR (70.1 ± 63.5 ng/ml; $t = 0.47$, df 28, $p = 0.64$). The *post hoc* comparison of the effects on the MATRICS-CCB obtained with the DMXBA-ER and DMXBA-IR found that the Neurocognitive Index T-scores were significantly elevated compared with the baseline for DMXBA-IR (4.20 ± 3.17 T-score units, $t = 5.31$, df 15, $p < 0.0001$). By comparison, the effect of DMXBA-ER was not significantly greater than baseline values (1.06 ± 6.26 T-score units). The most significant effect of DMXBA-IR had previously been found to be on the Attention-Vigilance Domain (Freedman *et al*, 2008). The effect of DMXBA-IR on this domain (7.80 ± 8.85 T-score units) was significantly greater than the effect of DMXBA-ER (-1.83 ± 6.46 T score units, $t = 3.61$, df 31, $p = 0.0011$; Figure 2). The placebo change in the Neurocognitive Index T-score with placebo was similar in the two trials (2.78 ± 5.98 T score units in the DMXBA-ER trial and 2.11 ± 6.27 in the DMXBA-IR trial; Table 2).

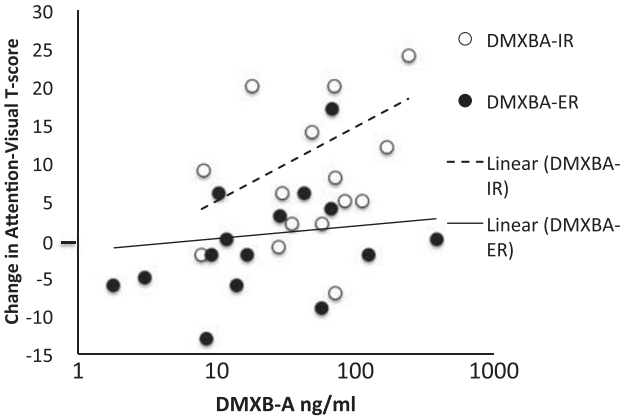

Figure 2 Comparison of treatment for 1 month with DMXBA-IR and DMXBA-ER formulation on the Attention-Vigilance Domain of the MATRICS-CCB in non-smoking schizophrenia patients. The effect of DMXBA-IR was significantly greater than the effect of DMXBA-ER (see text). The trendlines are illustrative only; neither is significant.

DISCUSSION

This is the first clinical trial that permits direct comparison of the effects of immediate-release and extended-release oral formulations of the same alpha7-nAChR agonist in schizophrenia. The cognitive effects observed with a brief-acting formulation were not seen with a longer acting formulation, despite nearly comparable plasma levels during testing. These results are consistent with the known physiological properties of alpha7-nAChRs, which display more extensive desensitization during prolonged exposure to agonists (Peng *et al*, 1994). They suggest that long duration-acting drugs are not likely to be effective for ameliorating neurocognitive deficits in schizophrenia.

The current trial demonstrates that even with a partial agonist, prolonged exposure to an alpha7-nAChR agonist is not an effective treatment strategy. A weak partial agonist like DMXB-A is an appropriate molecule for the investigation of pharmacokinetic limitations of alpha7-nAChR stimulation, because it is less likely to produce chronic desensitization than a stronger partial or full agonist that increases the probability that the activated receptors are converted into a stable desensitized state, rather than back into a resting state that can reactivated by agonist (Papke *et al*, 2009). Desensitization is intimately tied to receptor activation, so that full agonists have robust immediate effects, but they then produce more desensitization with loss of effect (Quick and Lester, 2002). Inverted U-shaped dose–response curves, indicative of desensitization-induced tachyphylaxis at higher agonist doses, have been observed in investigations of the effects of alpha7 agonists on cognitive and related prefrontal cortical functions in primates (Yang *et al*, 2013). Loss of pro-cognitive effect with chronic exposure to nicotine, a full agonist, is also well known. Although smoking produces short-duration bursts of very high nicotine levels, smokers' plasma levels of nicotine are chronically elevated. In earlier clinical tests with nicotine, we were unable to demonstrate any cognitive effect of acute nicotine, whether smoked or administered as nicotine gum, in schizophrenia patients who were chronic smokers, whereas acutely administered nicotine produced cognitive effects in non-smoking patients (Harris *et al*, 2004). Alpha7 receptors show some compensatory upregulation during chronic exposure to nicotinic agonists, but the effect is relatively small in comparison with the upregulation of alpha4–beta2 receptors (Lee *et al*, 2001).

In the current trial, the particularly low levels of DMXB-A in smokers were an additional confounding factor. Hepatic metabolism has been shown to be the limiting factor in the bioavailability of DMXB-A (Mahnir *et al*, 1998; Azuma *et al*, 1999). The metabolic profile for DMXB-A obtained *in vitro* shows predominantly *O*-demethylation at the 4-methoxy

Table 2 Comparison of Results From Trials of DMXBA-ER and DMXBA-IR in Non-Smokers and DMXBA-ER in Smokers, Means (SD)

Non-smokers	**DMXBA-ER baseline**	**DMXBA-ER 4 weeks**	**DMXBA-ER change**	**DMXBA-IR baseline**	**DMXBA-IR 4 weeks**	**DMXBA-IR change**
Number	18	18	18	15	15	15
MATRICS Neurocog. Comp T	34.17 (14.15)	35.22 (14.71)	1.06 (6.26)	35.93 (10.17)	40.13 (10.32)	4.20 (3.17)
MATRICS Attn-Vigil. T	37.44 (12.79)	35.61 (14.75)	−1.83 (6.46)	26.80 (8.50)	34.60 (7.69)	7.80 (8.85)
SANS	4.50 (2.46)	4.72 (3.03)	0.22 (1.56)	22.0 (18.3)	20.6 (18.3)	−1.4 (2.0)
BPRS	36.67 (8.20)	33.56 (6.52)	−3.11 (8.07)	30.0 (8.8)	27.4 (7.0)	−2.6 (4.9)
DMXBA (ng/ml)		56.2 (96.0)			70.1 (63.5)	
	Placebo-ER baseline	Placebo-ER 4 weeks	Placebo-ER change	Placebo-IR baseline	Placebo-IR 4 weeks	Placebo-IR change
Number	19	19	19	9	9	9
MATRICS Neurocog. Comp. T	39.72 (12.68)	42.50 (13.54)	2.78 (5.98)	41.44 (9.39)	43.55 (12.28)	2.11 (6.27)
MATRICS Attn-Vigil. T	40.39 (12.68)	39.78 (12.61)	−0.61 (5.38)	35.11 (13.11)	36.00 (11.77)	0.89 (8.91)
SANS	4.00 (3.14)	4.72 (3.63)	0.72 (1.81)	22.0 (18.3)	22.3 (18.8)	0.3 (2.4)
BPRS	34.33 (7.09)	32.17 (6.08)	−2.17 (4.63)	30.0 (18.8)	28.7 (9.9)	−1.3 (5.2)
Smokers	DMXBA-ER baseline	DMXBA-ER 4 weeks	DMXBA-ER change	Placebo-ER baseline	Placebo-ER 4 weeks	Placebo-ER change
Number	22	20	20	21	21	21
MATRICS Neurocog. Comp. T	32.00 (11.83)	32.29 (12.00)	0.29 (5.43)	31.14 (15.32)	32.67 (16.44)	1.52 (9.90)
MATRICS Attn-Vigil. T	32.57 (10.21)	33.33 (12.75)	0.76 (6.07)	30.00 (13.83)	34.71 (13.68)	4.71 (8.89)
SANS	5.30 (3.20)	4.80 (3.14)	−0.50 (2.48)	5.33 (2.73)	5.04 (2.99)	−0.29 (2.48)
BPRS	35.50 (10.60)	32.10 (8.58)	−3.40 (7.01)	37.90 (10.70)	33.19 (6.54)	−4.67 (8.75)
DMXBA (ng/ml)		4.2 (3.7)				
Nicotine (ng/ml)	18.6 (11.2)	19.8 (16.1)	1.2 (10.5)	20.1 (12.3)	14.0 (13.1)	−6.1 (18.4)
Cotinine (ng/ml)	106.6 (47.8)	113.7 (49.3)	7.2 (18.6)	135.1 (54.6)	122.2 (39.0)	−12.9 (50.6)

position of the benzylidene ring. This reaction is catalyzed by three different families of cytochrome P450 enzymes, including CYP1A1 and CYP1A2 (Azuma *et al*, 1999; Kem *et al*, 2004). Hepatic levels of these particular cytochrome P450 enzymes are increased by chronic cigarette smoking (Zevin and Benowitz, 1999), which may be the major explanation for the drastically reduced plasma levels of DMXB-A in the smoking group in our study.

The major finding relevant for future drug development comes from the exploratory *post hoc* comparisons with the results of the previously published study of the immediate-release formulation, using the newly available MCCB Neurocognitive Composite. The re-formulation of DMXB-A to obtain more extended levels led to the loss of such effects observed previously with the short immediate-release formulation. Plasma drug levels were determined immediately after MATRICS-CCB testing for both formulations, but the higher transient peak levels achieved by the faster release from the DMXBA-IR formulation and the subsequent rapid elimination are the likely reason for its greater effect. A limitation is that the comparison of DMXBA-ER and DMXBA-IR was not conducted in the same trial. However, the placebo response rates in the two trials, conducted at the same site, were quite similar. The comparison of the two formulations cannot fully distinguish between the alternative explanations that a high initial peak of activation is required, best achieved with immediate release of the agonist, compared with the possibility that the subsequent prolonged exposure from the extended-release formulation results in tachyphylaxis of the response.

Pharmaceutical companies that have developed agonist therapies for alpha7-nicotinic receptor in schizophrenia have selected drug candidates with long half-lives for once daily administration (Lieberman *et al*, 2013; Keefe *et al*, 2015; Haig *et al*, 2016a, 2016b). These drug candidates have produced promising results in initial, phase 2 trials, which have not been corroborated in the subsequent larger studies. DMXB-A-IR produced effects on the MATRICS-CCB Domains consistent with a meaningful clinical effect (Freedman *et al*, 2008). The increase of 4.20 ± 3.17 T-score units with DMXBA-IR is equivalent to the 4.8 ± 1.8 increase associated with employment in patients with schizophrenia, compared to patients who are not employed (August *et al*, 2012). Phasic stimulation may be particularly effective because DMXB-A activates a nitric-oxide-mediated second messenger response

in neurons that appears to extend its effect after brief-receptor stimulation (Adams *et al*, 2000).

A model for this interpretation is the cholinergic crisis or overstimulation produced by the extended presence of acetylcholine at neuromuscular nicotinic receptors in myasthenia gravis. Both myasthenia gravis and schizophrenia have in common a diminished expression of the relative nicotinic receptor, although the depletion is more severe in myasthenia (Freedman *et al*, 1995; Court *et al*, 1999; Guillozet-Bongaarts *et al*, 2014). The findings suggest that either judicious use of agonists, employing brief half-life drugs intermittently, or allosteric modulators to enhance the effects of phasically released acetylcholine may be required to achieve therapeutically significant cognitive effects through alpha7-nAChR receptor stimulation in schizophrenia (Gee *et al*, 2017).

FUNDING AND DISCLOSURE

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Supplementary Information accompanies the paper on the Neuropsychopharmacology website (http://www.nature.com/npp)