available at www.sciencedirect.com

ScienceDirect

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

Differential effects of ciproxifan and nicotine on impulsivity and attention measures in the 5-choice serial reaction time test☆

Mark Day[c], *Jia Bao Pan*[a], *Michael J. Buckley*[a], *Elizabeth Cronin*[a], *Peter R. Hollingsworth*[a], *Warren D. Hirst*[d], *Rachel Navarra*[d], *James P. Sullivan*[a], *Michael W. Decker*[a], *Gerard B. Fox*[a,b,*]

[a] *Neuroscience Research, Global Pharmaceutical Research and Development, Abbott Laboratories, Abbott Park, IL 60064, USA*
[b] *Advanced Technology, Global Pharmaceutical Research and Development, Abbott Laboratories, Abbott Park, IL 60064, USA*
[c] *Discovery Translational Medicine, Wyeth Research, 500 Arcola Road, PA 19426, USA*
[d] *Discovery Neuroscience, Wyeth Research, CN-8000, Princeton, NJ 08543, USA*

ARTICLE INFO

Article history:
Received 14 October 2006
Accepted 4 December 2006

ABSTRACT

Deficits in attention and response inhibition are apparent across several neurodegenerative and neuropsychiatric disorders for which current pharmacotherapy is inadequate. While it is difficult to model such executive processes in animals, the 5-choice serial reaction time test (5-CSRTT), which originated from the continuous performance test (CPT) in humans, may serve as a useful translational assay for efficacy in these key behavioral domains. At Wyeth and Abbott, we recently investigated the utility of employing the 5-CSRTT in adult rats. This involved training and testing groups of rats over an extended period of several months and required the animals to learn to nose-poke into one of five apertures following presentation of a brief visual stimulus in that aperture in order to obtain a food reward. When the stimulus duration was short, the rat had to pay close attention to make a correct choice—a nose-poke into the aperture with the brief visual stimulus. We evaluated nicotine and the histamine H_3 receptor antagonist, ciproxifan, since compounds targeting both nicotinic and histaminergic neurotransmission are currently under investigation for treating cognitive dysfunction in ADHD, AD and schizophrenia. After approximately 12 weeks of training, rats were tested with drug when they had achieved stable performance. Nicotine (0.2, 0.4 mg/kg s.c.) significantly improved accuracy and reduced errors of omission (reflecting improved attention and vigilance) when baseline performance was <90% correct. In contrast, nicotine tended to worsen accuracy when baseline performance was >90% correct. Using the same test paradigm, ciproxifan (3 mg/kg i.p.) reduced premature responding, a measure of impulsivity. Under conditions of variable stimulus duration, ciproxifan also improved accuracy and decreased impulsivity. In summary, we have replicated previous findings by others of positive effects of nicotine on attention, but also showed that this is dependent on baseline performance. We also expanded on previous positive findings by others with ciproxifan on attention and both Wyeth and Abbott demonstrate for the first time decreased impulsivity with this mechanism.

☆ This work was funded by Abbott Laboratories and Wyeth Research.
* *Corresponding author at*: Neuroscience Research, Global Pharmaceutical Research and Development, Abbott Laboratories, 100, Abbott Park Road, IL 60064, USA. Tel.: +1 847 937 8933; fax: +1 847 938 6025.
E-mail address: gerard.b.fox@abbott.com (G.B. Fox).
Abbreviations: 5-CSRTT, 5-choice serial reaction time test; CPT, continuous performance test; H_3R, histamine H_3 receptor; SD, stimulus duration; ITI, inter-trial interval; ADHD, attention deficit hyperactivity disorder; AD, Alzheimer's disease
0006-2952/$ – see front matter
doi:10.1016/j.bcp.2006.12.004

1. Introduction

Increased impulsivity and deficits in attention are key symptoms frequently observed across a number of different neuropsychiatric and neurodegenerative diseases [1–6]. Both symptoms are co-morbid in attention deficit hyperactivity disorder (ADHD), which is estimated to affect 6–10% of school-aged children [7], and in schizophrenia, where poor functional outcome in the majority of patients is predicted by such cognitive and behavioral deficits [8]. In both ADHD and schizophrenia, these deficits ultimately lead to severe impairment of executive functioning, which remains largely untreated. To add to the complexity, the word 'attention' itself is an umbrella term for a collection of distinct executive processes, such as selective and divided attention, vigilance, and distractibility. In addition, impulsivity, which may be defined as the failure of response inhibition, has further been linked to increased probability of suicide, gambling, drug abuse and aggression. Indeed, schizophrenic patients with a history of substance abuse and addiction exhibit higher levels of sensation seeking and impulsivity [9] and are a high risk for suicide [10,11]. Consequently, there is much interest in modeling executive function preclinically to help identify new pharmacological therapies for ADHD and schizophrenia.

The continuous performance test (CPT) has been widely used for over 50 years to measure attention performance in humans [12]: it is sensitive in detecting attention deficits across several disorders such as mild cognitive impairment (MCI) [13], schizophrenia [14–17] and ADHD [1,18–20]. In the CPT, subjects are required to respond to a specific visual stimulus (e.g. the letter "X") presented on a visual display unit: this stimulus has a much lower probability of appearing than other stimuli (e.g. A, C, F, M). Since "X" occurs with less frequency, the ability to respond with a high percentage of correct responses requires the subjects to remain attentive during the trial types. When the subjects see the letter "X" they are required to press a button/click a computer mouse. This simple response affords several measures to be taken from the subject regarding attentional accuracy (correct response on the button/computer mouse), false alarm hit rate (number of errors or responses when a letter other than "X" is presented), processing speeds (latency to press the button or click on the mouse when presented with "X") and impulsivity measures (making a response prior to the presentation of a stimulus). There are now several versions of the CPT, some of which manipulate test parameters in order to increase the attentional load. For example, in the "AX" version of the CPT [14], the subject is required to respond to X only when it is preceded by the letter "A". On the CPT, ADHD children show overall lower scores as measured by increased impulsive and incorrect responding. Moreover, these effects can be reversed with the stimulant methylphenidate (Ritalin®) [21,19]. The CPT is also used as an attention assay in the 'measurement and treatment research to improve cognition in schizophrenia' (MATRICS) test battery to evaluate the efficacy of pro-cognitive drugs in clinical trials in schizophrenic patients [22–24]. However, stimulant based drugs that are used to treat attention deficits, such as amphetamine, caffeine, and methylphenidate, may not be suitable pro-attentive therapies for schizophrenia, since some have abuse potential and may even aggravate symptoms [25].

The preclinical analogue of the CPT is the 5-choice serial reaction time test (5-CSRTT) [12]. In this operant-based test, which was originally developed by Robbins [12] from the CPT and has been widely characterized, animals are required to be attentive and withhold responding while monitoring five apertures for a brief (e.g. 1 s or less) illumination behind one of these apertures. Thus, in the 5-CSRTT in rats, the apertures are the equivalent of non-target letters in the CPT in humans; brief light stimuli are analogous to the presentation of the letter "X" in the CPT in humans. While human subjects watch for the "correct" letter and push a button/click a computer mouse, rats performing the analogous task in the 5-CSRTT must nose-poke into the correct aperture (where the brief light stimulus was just presented) to receive a food reward. The same behavioral measures (accuracy, omissions, premature responding, speed of response) can be recorded and quantitated.

While stimulants that are used to treat ADHD such as methylphenidate exert positive effects in the 5-CSRTT, one of the best-studied pharmacological effects is with nicotine, particularly at the labs of Stolerman and colleagues [26]. In serial preclinical studies in rats, acute nicotine administration increased response accuracy, reduced omission errors and decreased reaction time [12,26]. Interestingly, these investigators also showed improved attention in the 5-CSRTT following repeated administration of nicotine. Concurrent with this, animals exposed to repeated nicotine treatment also demonstrated a decrease in correct response latency and an increase in anticipatory responding, implying a link between nicotine's locomotor stimulant effects and an increase in impulsive-like behavior. Clearly, depending on how nicotine is administered, differential activity can be observed in the 5-CSRTT, sometimes making interpretation difficult. This is important since several selective agonists targeting $\alpha 4\beta 2$ neuronal nicotinic receptors (e.g. ABT-089, TC-1734) are already progressing through clinical development and offer great potential for translational studies.

Histaminergic neurotransmission is also the subject of intense investigation in recent years and histaminergic H_3 receptors (H_3Rs), which are constitutively active and highly expressed in the central nervous system (CNS) as auto- and heteroreceptors, have been implicated in sleep [27–32], arousal [28,31,33,34], information processing and cognition [32,35–41]. In addition to direct regulation of histamine release, H_3Rs also directly regulate cholinergic neurotransmission and can modulate the release of dopamine, serotonin and norepinephrine [30]. However, the effects of H_3R ligands are much less well studied in the 5-CSRTT. A limited early study with ciproxifan did demonstrate an effect on attention measures, observed when demands on attention were increased [42]. The only other published 5-CSRTT study with a H_3R ligand that we could find demonstrated a failure of the antagonist, thioperamide, to reverse a scopolamine-induced attention deficit [43]. Thus, there is a large gap in our ability to translate efficacy in rats using 5-CSRTT to CPT in humans for this mechanism and there are limited translational data available for nicotinic agonists in the same regard. In addition, it is not clear from the existing literature whether pro-cognitive efficacy resulting from blockade of H_3Rs with specific antagonists or from

selective activation of nicotinic receptors in other behavior models of ADHD is mediated through effects on attention or impulsivity or both. As a consequence, we sought to further characterize the effects of nicotine and ciproxifan in the 5-CSRTT in adult rats. Independent data sets originating from Neuroscience Departments at both Wyeth Research and Abbott Labs were compiled and we now present new data that corroborate differential effects of nicotine and ciproxifan on separate measures of attention and impulsivity under similar test conditions.

2. Methods

2.1. Animals and housing

Adult, male hooded Lister rats (Harlan, UK), weighing 350–400 g prior to training were used at Abbott while adult male Long Evans rats (Portage, MI, USA) were used at Wyeth. All rats had free access to water and were placed on a food-restricted diet to maintain them at 85% of their free-feeding body weights. Animals were single housed under a 12 h light:12 h dark schedule (lights on at 06:00 a.m.). All studies were conducted in the light phase and in accordance with Abbott and Wyeth Animal Care and Use Committee and National Institutes of Health Guide for Care and Use of Laboratory Animals guidelines in facilities accredited by the Association for the Assessment and Accreditation of Laboratory Animal Care.

2.2. Drugs

Ciproxifan, which was purchased (Wyeth) from Sigma–Aldrich (St. Louis, MO, USA) or synthesized at Abbott (Abbott Park, IL, USA), was dissolved in 2% Tween 80® in water. Nicotine was purchased from Sigma–Aldrich (St. Louis, MO, USA) and dissolved in saline. Sterile saline was purchased from Abbott Labs (Abbott Park, IL, USA). Ciproxifan was administered intraperitoneally (i.p.) while nicotine was administered subcutaneously (s.c.) in an injection volume of 1 mL/kg. Saline vehicle was administered i.p. or s.c. using an injection volume of 1 mL/kg.

2.3. 5-CSRTT apparatus

The 5-CSRTT apparatus consists of eight aluminum operant chambers (26 cm × 26 cm × 26 cm), purchased from Lafayette Instruments Ltd. Lafayette, IN, USA (Abbott Laboratories) or ten aluminum chambers (25 cm × 25 cm × 25 cm) purchased from Med Associates Inc., St. Albans, VT, USA (Wyeth Research). Each of the chambers (see Fig. 1A for an example from Abbott) is individually housed within sound-attenuating cabinets and is ventilated by low-level noise fans, which also serve to mask extraneous background noise. The concave-curved rear wall of each chamber contains five 2.5 cm square receptacles 4 cm deep positioned 2 cm above floor level (Fig. 1B). The entrance of each hole is equipped with a photocell, which monitors interruptions of a beam of infrared light as a result of nose pokes; a standard light-emitting diode (LED) is located at the rear of the hole for presentation of visual stimuli. A food magazine dispenser equipped with a flap and an infrared or micro switch nose-poke detector at the entrance is located on the opposite wall equidistant from each hole (Fig. 1A and B). Each chamber is illuminated with a house light mounted on the ceiling. The apparatus and data collection at Abbott are controlled by Cambridge software written in Turandot (Cambridge Cognition Limited, Sileby, Loughborough, UK) while those at Wyeth are controlled by software developed by Conclusive Solutions (Harlow, UK).

2.4. 5-CSRTT training

Briefly, before entering different training sessions, animals were trained first to learn two major behavioral components involved in the 5-CSRTT, nose-poking at an illuminated receptacle and obtaining food from the magazine. During multiple training sessions over 12 weeks, animals learn to nose-poke in response to randomly illuminated receptacles (by the LED). After nose-poking into the illuminated receptacle (Fig. 1C), the animal turns around to obtain a food reward (pellet) from the food magazine (Fig. 1D). Gradually, the duration in which the LED illuminates any given receptacle, the stimulus duration (SD), is decreased until the rat must maintain attention and vigilance in order to detect the stimulus to obtain the food reward. The interval between each trial is called the inter-trial interval (ITI) and is usually set between 5 and 15 s. When animals had been trained to perform at a level of at least 75% correct (i.e. 75% correct choices for the aperture with the visual stimulus) on the standard procedure (1 s SD and 5 s inter-trial interval (ITI) at Abbott or 0.5 s SD and 5 s ITI at Wyeth), testing could commence.

2.5. 5-CSRTT behavioral measures assessed

The 5-CSRTT allows measurement of a number of distinct behaviors that translate to the human equivalent in the CPT. In the current 5-CSRTT studies, we were particularly interested in assessing attention and impulsivity measures as well as speed of processing.

2.5.1. Attention

To measure attention, the percent of trials that resulted in a correct nose-poke to the visual stimulus presented in a particular aperture can be converted into a % correct measure of accuracy, which is dependent on intact attention, particularly under conditions where the visual stimulus duration is short. This behavior was measured at both Abbott and Wyeth. In addition at Abbott, the number of missed trials (or omission errors) where the rat failed to respond at all to a presented stimulus was recorded as another measure of attention.

2.5.2. Impulsivity

To measure impulsivity, the number of anticipatory (or premature) responses was recorded at both Abbott and Wyeth: this is defined as the number of times each rat nose-poked between trials (i.e. during the ITI) in the absence of a visual stimulus in any of the apertures. Thus, if the animal is unable to withhold responding this will be represented as an increase in impulsivity. A basal level of premature responding is normal so it is usually possible to detect both an increase or a decrease in impulsive behavior.

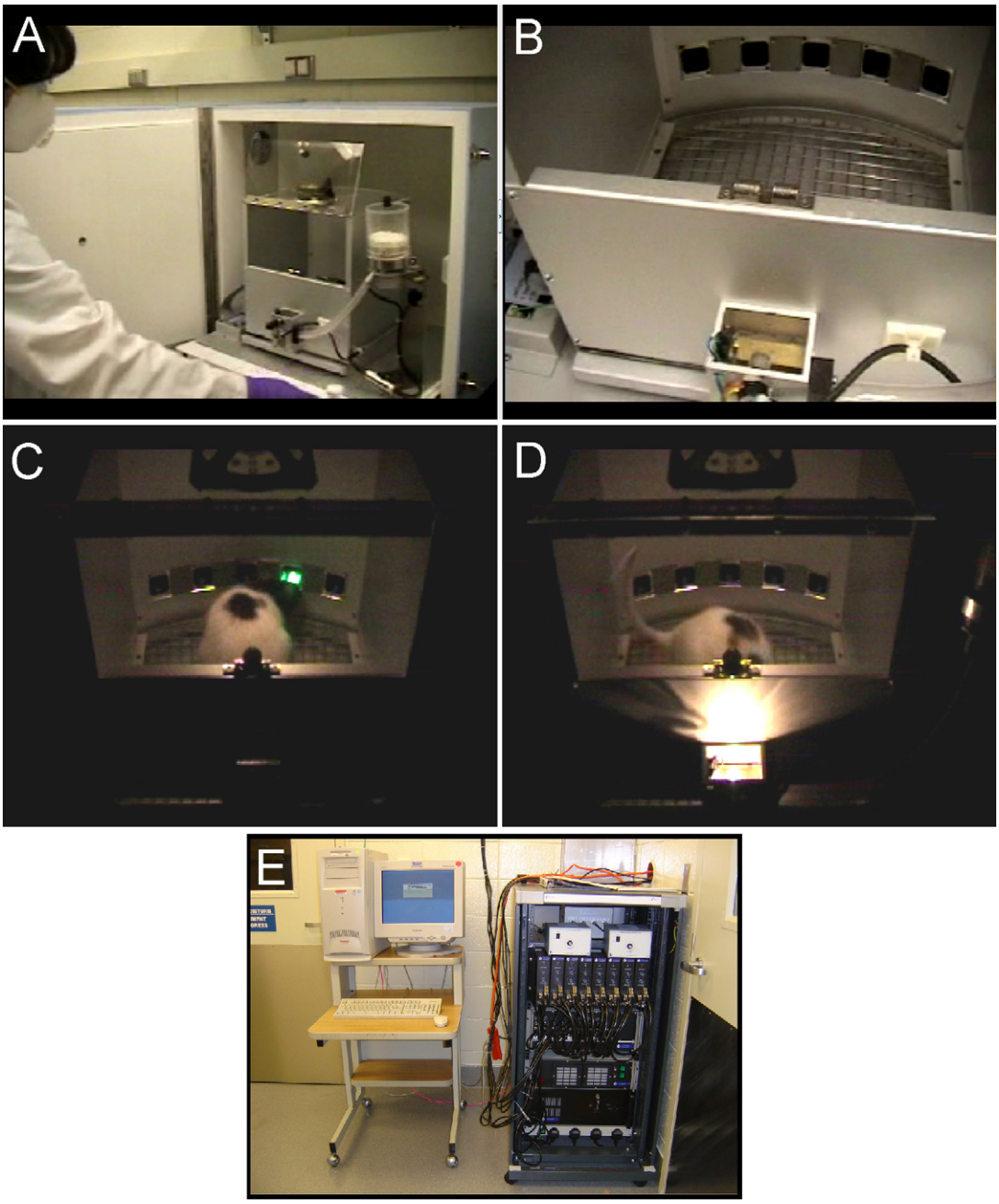

Fig. 1 – Typical 5-CSRTT equipment at Abbott Laboratories. Shown is a single test apparatus located within a sound-attenuating cabinet. For illustrative purposes, the experimenter has opened the door of the cabinet (A) as well as the test chamber within. Visible to the right hand side of the test chamber is the food pellet dispenser, which is connected to the food magazine on the front of the apparatus for delivery to the rat by a short length of clear tubing. Recessed within the rear, curved wall (B) are nine apertures, four of which are blocked leaving five apertures open for 5-CSRTT testing. Also visible is the wire mesh floor for easy cleaning (B). With the door to the sound attenuating cabinet remaining open for illustrative purposes (normally this would be closed) and only the test chamber house light on, a rat can be seen making a correct choice by nose-poking into the aperture second from the right following illumination with a LED located at the back of that aperture (C). This nose-poke breaks an optical beam within the floor of the aperture triggering the release of one food pellet from the food pellet dispenser into the magazine tray, which is illuminated by a filament lamp on the opposite wall (D; behind the rat). The rat turns to retrieve the food pellet and quickly reorients to face the curved wall containing the five apertures to await the next short, visible stimulus. Task parameters can be controlled for varying levels of difficulty. In the current studies, the duration of the stimulus and the time between each stimulus presentation were manipulated to increase attentional demand and assess impulsivity. The entire apparatus (8 test chambers at Abbott and 10 test chambers at Wyeth) has a dedicated interface and is computer controlled (E).

2.5.3. *Speed of processing*

To measure how rapidly a rat responds to the visual stimulus, the latency to nose-poke to the visual stimulus in the correct aperture was recorded at both Abbott and Wyeth. This information can be used to assess whether the drug treatment(s) had any effect on general motorfunction.

2.6. *Data analysis*

A within-subject design was used in all experiments, in which each animal received each treatment once in a randomized order. Animals were tested twice weekly and trained on other weekdays to reinstate baseline performance; a minimum 2 days washout period was allowed between test days. All tests lasted 30 min (Abbott) or 100 trials (Wyeth).

Outcome measures were:

(1) Percent correct response = [correct responses/(correct + incorrect responses)] × 100.
(2) Percent of omission errors = (omission errors/number of stimuli presented) × 100.
(3) Anticipatory response rate = (number of responses in ITI/number of trials)/ITI length (s).
(4) Premature responses = (number of responses in ITI/number of trials)
(5) Latency of correct responses = the mean time between stimulus onset and a nose poke in the correct hole.

To maximize normal distribution and homogeneity of variances for statistical analyses, percentage data were arcsine transformed, latency data were log transformed and anticipatory response data were square root transformed as per Stolerman et al. [16]. Results in the figures are presented as raw values, however. All transformed data were analyzed by one-way repeated measures ANOVA followed by Tukey's multiple comparison tests if the one-way repeated measures ANOVA showed significance ($p < 0.05$). Prism III and IV (GraphPad software, Inc. USA) and Microsoft Excel (Microsoft Corporation, WA, USA) were used for all data plotting and analyses.

3. Experimental design and results

3.1. *Abbott laboratories: effect of nicotine*

Two separate experiments using a total of 15 rats in each case were designed to assess the effects of nicotine on attention, impulsivity and processing speed.

Experiment 1. All animals in the first experiment were drug naïve, except for pretreatement with nicotine (0.2 mg/kg s.c.)

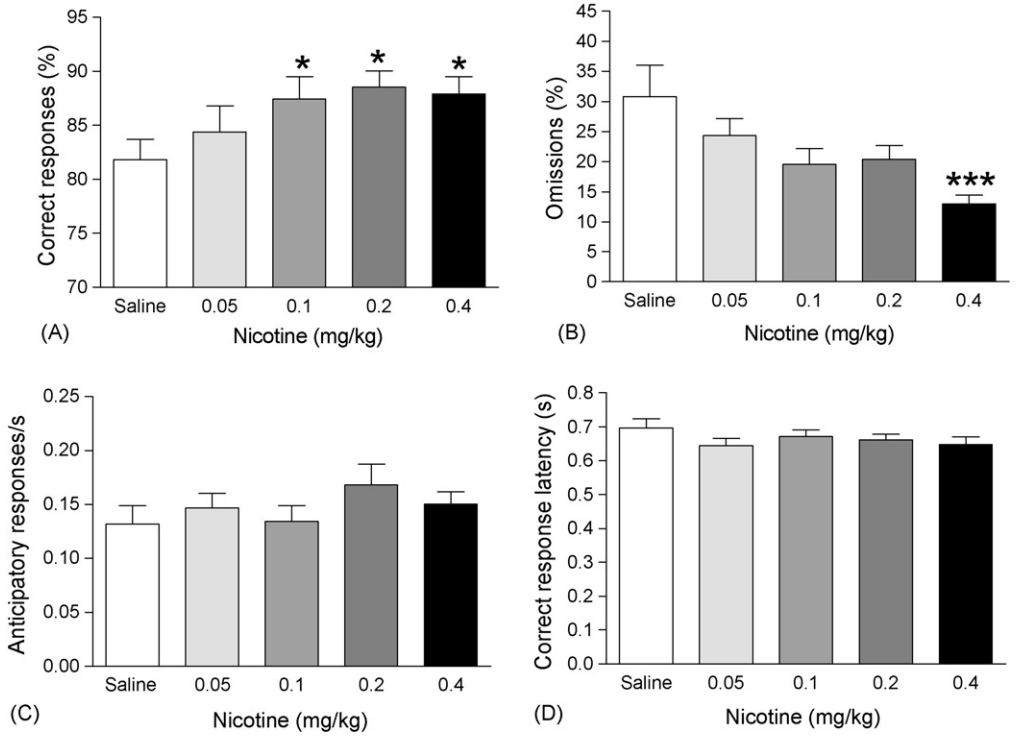

Fig. 2 – Effect of acute nicotine administration on performance of rats in the 5-CSRTT. Data are from experiment 1, conducted at Abbott Laboratories. Rats were naïve to any treatment at time of testing. Our statistical analyses conducted on subjects with lower baseline performance (percent correct response <90%) showed that nicotine significantly ($F[4,36] = 6.164$, $p = 0.0007$) enhanced correct responding (A) and decreased ($F[4,36] = 5.498$, $p = 0.0015$) omission errors (B). No effect ($F[4,36] = 1.345$, $p = 0.2723$) was observed on anticipatory responding (C), nor ($F[4,36] = 2.564$, $p = 0.0548$) on correct response latency (D). Animals were tested under conditions of SD = 1 s and ITI = 15 s, $n = 15$ per study. Data are shown for $n = 10$ rats with baseline responding <90% correct (cut-off point). Results are expressed as mean ± S.E.M. Data were analyzed by one-way repeated measures ANOVA followed by Tukey's multiple comparison test if one-way repeated measures ANOVA showed significance ($p < 0.05$). $^{*}p < 0.05$ and $^{***}p < 0.001$ compared to saline treatment group.

daily at 2 h after training for 3 consecutive days during the week prior to commencing testing. This was done in advance of testing in order to habituate the rats to any potential non-specific effects of the drug. Rats were trained to perform the task under conditions of SD = 1 s and ITI = 5 s and tested under conditions of SD = 1 s and ITI = 15 s. In the test sessions, animals were administered vehicle or nicotine at 0.05, 0.1, 0.2 or 0.4 mg/kg, s.c., 10 min prior to the test. Baseline percent correct response measures were high in some rats (>90%; $n = 5$), given the relatively long (1 s) stimulus duration during testing. Our statistical analyses (see Fig. 2 legend) confirmed that nicotine did not improve attention in those animals with baseline responding >90%. In contrast, the same analysis conducted on subjects with lower baseline performance (percent correct response <90%; $n = 10$), showed that nicotine dose-dependently enhanced correct responding (Fig. 2A) and decreased the overall number of trials that were missed (Fig. 2B), effects indicative of increased attention and vigilance. However, nicotine had no significant effect on either anticipatory responding (Fig. 2C), a measure of impulsivity, nor latency to make a correct response (Fig. 2D), a measure of processing speed. These rats were reused in further experiments and a 3-week drug washout period was allowed between each study. During the washout period, animals received training every weekday.

Experiment 2. In the second experiment animals were previously tested in either experiment 1 (above) or experiment 4 (below). Rats were trained to perform the task under conditions of SD = 0.6 s and ITI = 5 s and tested at under conditions of SD = 0.6 s and ITI = 15 s. All animals were also pretreated with nicotine (0.4 mg/kg s.c.) daily at 2 h after training for 3 consecutive days during the week prior to commencing testing in order to habituate to any potential non-specific effects of the drug. Our statistical analyses (see Fig. 3 legend) conducted for rats with correct responding <90% revealed that nicotine again enhanced measures of attention and vigilance by increasing correct responding (Fig. 3A) and decreasing the number of trials missed (Fig. 3B) (see Fig. 3 legend for statistical analyses). Interestingly, nicotine produced an increase in impulsivity at all doses tested (see Fig. 3C). However, no effects were observed on processing speeds utilizing reaction time measures (Fig. 3D).

3.2. *Abbott laboratories: effect of ciproxifan*

Two separate experiments using a total of 12–13 rats in each case were designed to assess the effects of ciproxifan on

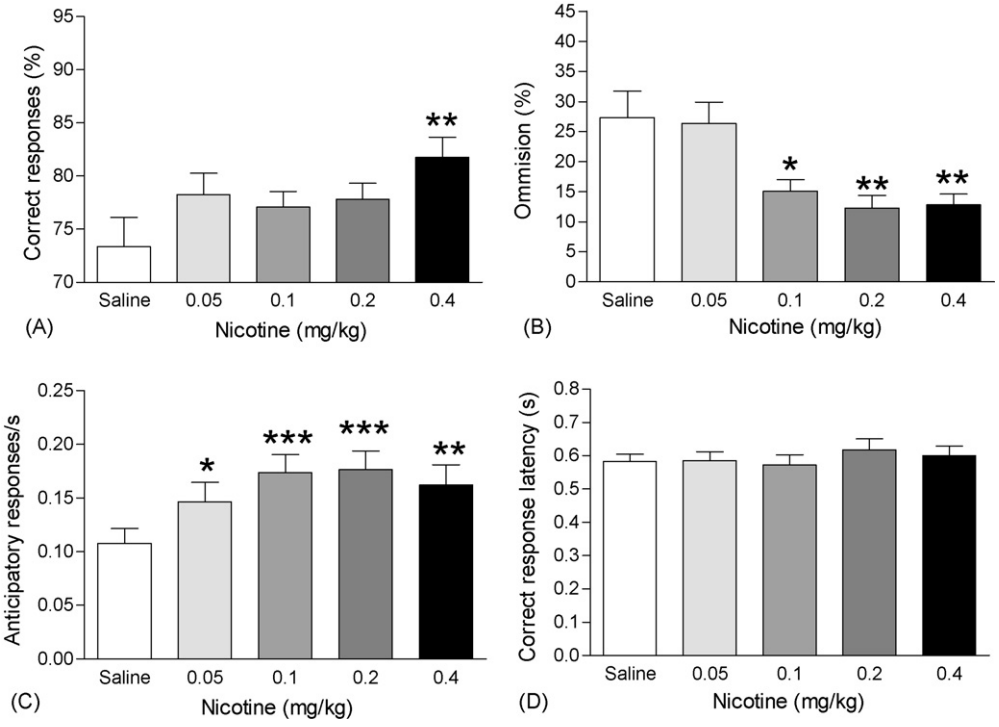

Fig. 3 – Effect of nicotine on performance of rats in the 5-CSRTT. Data are from experiment 2, conducted at Abbott Laboratories. Rats were previously tested with nicotine (same rats from Fig. 2) or with ciproxifan (same rats from Fig. 5) so a 3-week washout period was allowed. Nicotine significantly ($F[4,48] = 3.437$, $p = 0.015$) increased correct responding indicative of improved attention (A) and significantly ($F[4,48] = 57.806$, $p < 0.0001$) decreased omissions indicative of improved vigilance (B). In this experiment, nicotine also significantly ($F[4,48] = 8.221$, $p < 0.0001$) increased anticipatory responding (C) indicating that re-exposure to nicotine may have produced an impulsive-like behavior that was independent ($F[4,48] = 0.8148$, $p = 0.5220$) of any non-specific changes in motor function (D). Animals were tested under conditions of SD = 0.6 s and ITI = 15 s (SD was shorter to maintain stable baseline performance), $n = 15$ per study. Data are shown for $n = 13$ rats with baseline responding <90% correct (cut-off point). Results are expressed as mean ± S.E.M. Data were analyzed by one-way repeated measures ANOVA followed by Tukey's multiple comparison test if one-way repeated measures ANOVA showed significance ($p < 0.05$). $^{*}p < 0.05$, $^{**}p < 0.01$ and $^{***}p < 0.001$ compared to saline treatment group.

attention, impulsivity and processing speed. The same rats described in Experiment 1 were used again after a 3-week washout.

Experiment 3. During the washout period, rats were trained under conditions of SD = 1 s and ITI = 5 s. In test sessions, ITI was 15 s and SD was 1 s (the same conditions used for nicotine in experiment 1). Animals were treated with vehicle or ciproxifan at 0.3, 1 or 3 mg/kg, i.p., 1 h prior to the commencement of testing. Animals were injected with saline (1 mL/kg, i.p.) daily at 2 h after training for 3 consecutive days during the week prior to commencing testing in order to habituate to any non-specific effects of i.p. injections. Our statistical analyses (see Fig. 4 legend) revealed that ciproxifan significantly decreased impulsivity as measured by anticipatory responses (Fig. 4C). In contrast, no significant effects were observed on measures of attention (Fig. 4A), vigilance (Fig. 4B) or processing speeds utilizing reaction time measures (Fig. 4D).

Experiment 4. A different set of 12 naïve animals was used in experiment 4. Animals were treated with vehicle or ciproxifan at 0.3, 1 or 3 mg/kg, i.p., 1 h prior to the commencement of testing. Rats were also pretreated with ciproxifan (3 mg/kg, i.p.) daily at 2 h after training for 3 consecutive days during the week prior to commencing testing in order to habituate to any potential non-specific effects of the drug. Animals were trained under conditions of SD = 0.5 s, ITI = 5 s and tested at SD = 0.25 s and ITI = 5 s. When tested under these challenging conditions, ciproxifan again significantly (see Fig. 5 legend) decreased impulsivity as measured by anticipatory responses (Fig. 5C). Similar to experiment 3 for ciproxifan above, under the current test conditions ciproxifan still failed to show any improvement on measures of attention (Fig. 5A) or vigilance (Fig. 5B), despite the larger window created by the shorter SD (Fig. 5A). Ciproxifan also had no effect on processing speeds utilizing reaction time measures (Fig. 5D).

3.3. *Wyeth research: effect of ciproxifan*

This experiment, using a total of 15 rats, was designed to assess the effects of ciproxifan on attention, impulsivity and processing speed. In particular, attentional load was increased by introducing variable stimulus durations.

Experiment 5. Animals were trained to perform at a level of 75% correct on the standard procedure (SD = 0.5 s, ITI = 5 s) following which drug testing began using a variable SD schedule (SD = 0.5, 0.2, 0.1 or 0.01 s). In these test sessions, animals were administered vehicle or ciproxifan at 1, 3, or 10 mg/kg, i.p. 30 min prior to the test. Equal numbers of each of the 4 SDs were randomly presented during the 100 trial session. Under the most demanding conditions of a variable SD (0.5, 0.2, 0.1, 0.01 s), for vehicle treatment, our statistical analyses confirmed that as the stimulus duration decreased attention became increasingly impaired, confirming that each

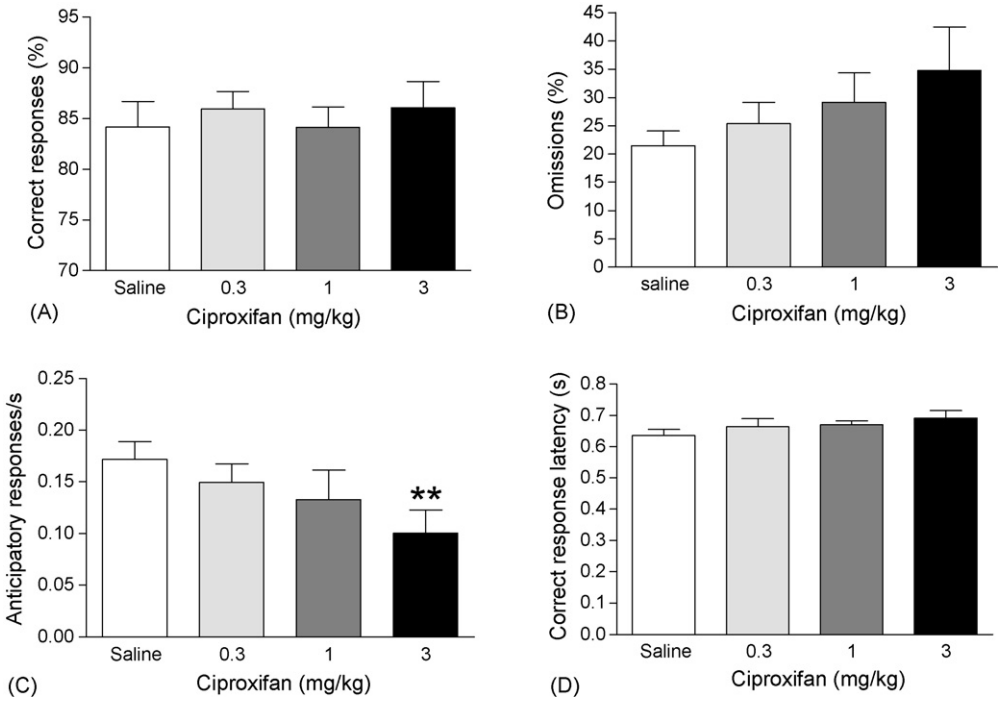

Fig. 4 – Effect of acute ciproxifan administration on performance of rats in the 5-CSRTT. Data are from experiment 3, conducted at Abbott Laboratories. Rats were previously tested with nicotine (same rats from Fig. 2) so a 3-week washout period was allowed. No significant effect ($F[3,33] = 0.5500$, $p = 0.5847$) on correct responding (A) or omissions ($F[3,33] = 1.629$, $p = 0.2014$) (B) indicated no effects on measures of attention or vigilance. Anticipatory responding (C) was significantly decreased ($F[3,33] = 5.235$, $p = 0.0046$) indicative of reduced impulsivity that was independent ($F[3,33] = 0.2.255$, $p = 0.1003$) of any non-specific changes in motor function (D). Animals were tested under conditions of SD = 1 s and ITI = 15 s, $n = 12$ per study. Data are shown for all $n = 12$ rats. Results are expressed as mean ± S.E.M. Data were analyzed by one-way repeated measures ANOVA followed by Tukey's multiple comparison test if one-way repeated measures ANOVA showed significance ($p < 0.05$). $^{**}p < 0.01$ compared to saline treatment group.

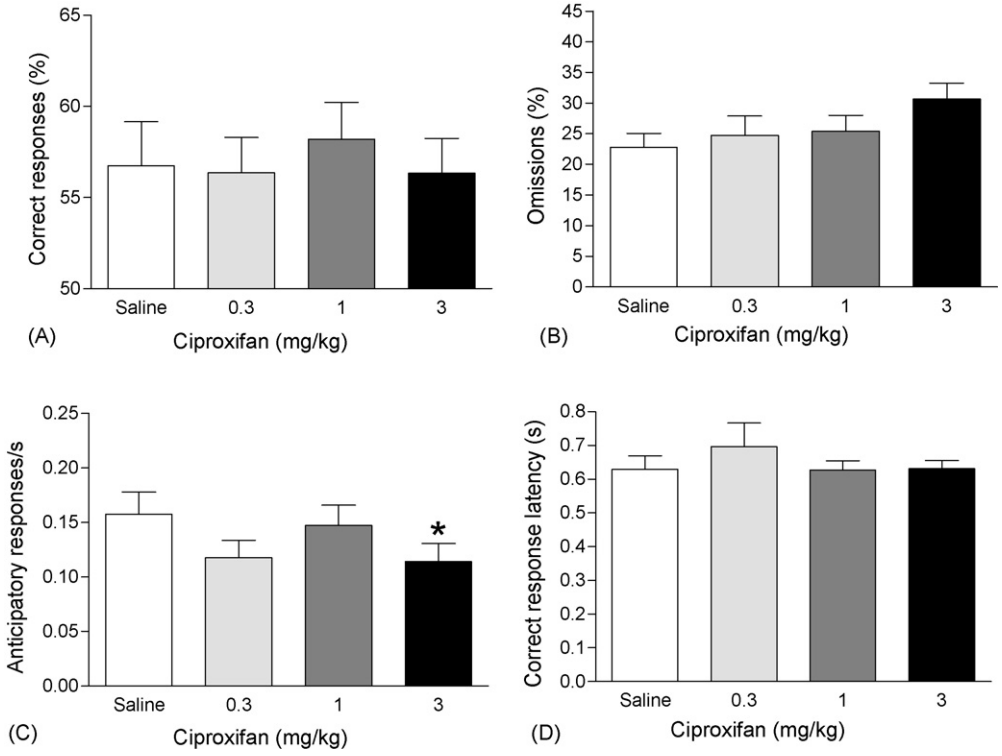

Fig. 5 – Effect of acute ciproxifan administration on performance of rats in the 5-CSRTT. Data are from experiment 4, conducted at Abbott Laboratories. Rats were naïve to any treatment prior to testing. No significant effect ($F[3,42] = 0.5945$, $p = 0.6221$) on correct responding (A) nor on omissions ($F[3,42] = 1.871$, $p = 0.1491$) (B) indicated no effects on measures of attention or vigilance. Similar to experiment 3, anticipatory responding was significantly ($F[3,42] = 4.112$, $p = 0.0121$) decreased (C) indicative of reduced impulsivity that was independent ($F[4,42] = 0.5983$, $p = 0.6197$) of any non-specific changes in motor function (D). Animals were tested under conditions of SD = 0.25 s and ITI = 5 s, $n = 15$ per study. Data are shown for all $n = 15$ rats. Results are expressed as mean ± S.E.M. Data were analyzed by one-way repeated measures ANOVA followed by Tukey's multiple comparison test if one-way repeated measures ANOVA showed significance ($p < 0.05$). $^{*}p < 0.05$ compared to saline treatment group.

SD resulted in a significant decrease in correct responding from the next (not shown). Moreover, ciproxifan significantly (see Fig. 6 legend) improved attention as measured by correct responding and shown for two different SDs (Fig. 6A). In agreement with findings at Abbott where ciproxifan decreased impulsivity, at Wyeth ciproxifan also significantly decreased impulsivity (Fig. 6B) when attentional demand was high. Ciproxifan did not affect processing speed (Fig. 6C). An analysis of the number of missed trials (not shown) also revealed a significant main effect of stimulus and a main effect of dose that was attributable to the highest dose (10 mg/kg) of ciproxifan increasing the number of missed trials at SD = 0.01 s only (not shown).

4. Discussion

The 5-CSRTT has been used to investigate the effects of systemic administration of many drugs as well as the effects of specific neurochemical lesions on various aspects of attention that are relevant for neurological disorders (for a thorough review, see [12]) such as schizophrenia and ADHD. Indeed, the 5-CSRTT was originally developed to increase our understanding of the nature of the behavioral deficits observed in children with ADHD. However, this test is typically difficult to operate in a drug discovery environment as animals require training for many weeks in order to achieve an acceptable baseline performance level. Since drug discovery scientists require high test–retest reliability and the ability to evaluate many novel drugs, it is often beneficial for scientists to combine resources to understand new mechanisms. Thus, investigators at Abbott Laboratories and at Wyeth Research pooled data from independent 5-CSRTT studies of reference compounds from two mechanisms, cholinergic and histaminergic, currently of interest for developing new drugs for ADHD.

In the present studies, we demonstrate significant improvements in measures of attention and vigilance (improved accuracy and decreased errors of omission) following administration of the cholinergic agonist, nicotine, to rats. These improvements were observed with acute treatment as well as in rats that had been previously exposed (and arguably, sensitized) to nicotine, although with previous exposure, there was also an increase in premature responding. The histamine H_3R antagonist, ciproxifan, also improved attention, but only under conditions at Wyeth where attentional demand was highest with the use of variable stimulus

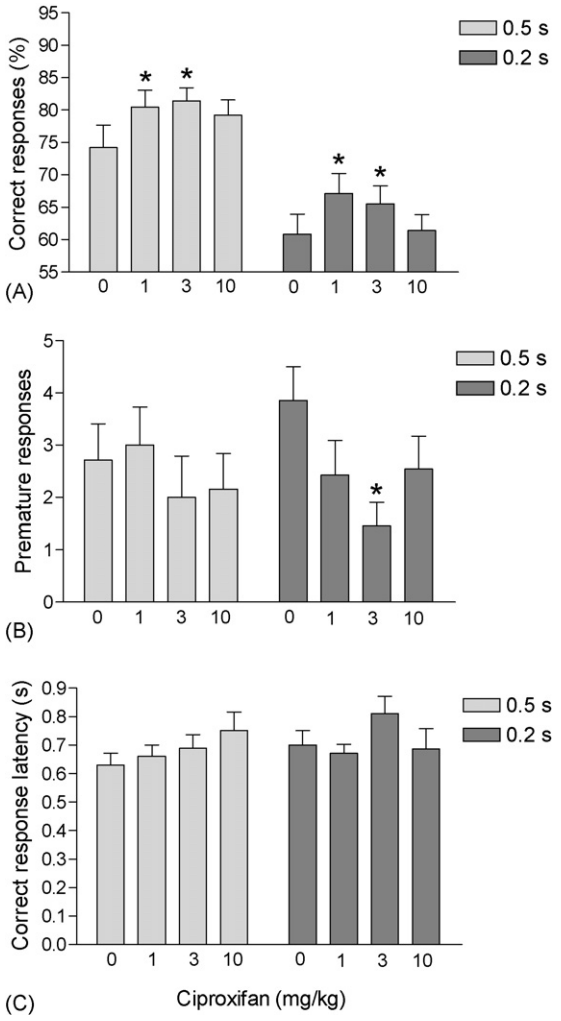

Fig. 6 – Effect of acute ciproxifan administration on performance of rats in the 5-CSRTT. Data are from experiment 5, conducted at Wyeth Research. Rats were drug free prior to testing. A significant ($F[3,104] = 5.235$, $p = 0.05$) effect on percent correct responding (A) and a significant ($F[3,208] = 3.43$, $p < 0.01$) effect on anticipatory responding (B) indicated that attention was enhanced and impulsivity decreased and that these effects were independent ($F < 1$) of any non-specific changes in motor function (C). Animals were tested under conditions of SD = 0.5, 0.2, 0.1, 0.01 s and ITI = 5 s, $n = 15$. A significant main effect of stimulus duration ($F[2,208] = 115$, $p < 0.001$) was also observed, confirming that each SD resulted in a significant decrease from the next. Data are shown for all $n = 15$ rats. Results are expressed as mean ± S.E.M. Data were analyzed by one-way repeated measures ANOVA followed by Tukey's multiple comparison test if one-way repeated measures ANOVA showed significance ($p < 0.05$). *$p < 0.01$ compared to vehicle treatment group.

durations. In contrast, at Abbott, where stimulus duration did not change, ciproxifan did not improve attentional measures. However, of most interest, ciproxifan also decreased measures of impulsivity at both Abbott and Wyeth, significantly decreasing premature responding. These latter effects on impulsivity have not been reported previously for this mechanism, were not apparent with nicotine, and thus clearly differentiate histaminergic H_3R antagonism from nicotinic receptor agonism on behavioral measures relevant for ADHD.

4.1. *Nicotine studies*

Our 5-CSRTT data with nicotine are consistent with previous findings demonstrating dose-related improvements in attention with this drug [19,26,44–47]. In the present study, nicotine significantly increased accuracy and reduced omission errors in rats with a baseline performance of 90% correct or less. Interestingly, rats with higher baseline performance levels were not improved: rather a trend toward a decrease in performance was evident. While this latter trend has not been previously reported it is interesting to note that nicotine has been shown to have a greater effect in Sprague-Dawley rats compared to Hooded Lister rats. This is relevant since baseline 5-CSRTT performance in Sprague-Dawley rats tends to be lower than for hooded Lister rats [26,45]. Thus, for cholinergic compounds such as nicotine, baseline performance levels at the commencement of pharmacologic testing in the 5-CSRTT should be carefully controlled.

Nicotine, at all doses tested, had no significant acute effect on either latency or anticipatory responding, indicating that the decrease in omission rate after acute dosing was not due to non-specific motor or motivation effects. These data also further confirm that the increase in accuracy after acute treatment was not related to any stimulant-like activity of nicotine. However, a stimulant effect of nicotine was demonstrated clearly in a second experiment, where rats that had been previously exposed to nicotine exhibited a significant increase in anticipatory responding. Despite this, however, the magnitude of the improvement of attentional measures remained similar. Repeated administration of nicotine can induce behavioral sensitization or tolerance to various measures including locomotor function [48,49]. Hahn et al. demonstrated that chronic (6 weeks) nicotine treatment significantly increased anticipatory responding and decreased response latency [44], effects that are consistent with potential sensitization. In the present study, repeated exposure to nicotine resulted in a significant increase in anticipatory responding but had no effect on response latency, thus implying a disassociation between increased anticipatory responding and possible response bias as a consequence of increased motor activity. It is also important to note that anticipatory responding reflects activity during the ITI with no reward consequences. Thus, an increase in anticipatory responding is not related to any motivational components of the task, but rather may be considered as a disinhibition behavior reflecting impulsive activity. Olausson and colleagues have also reported that behavioral sensitization to nicotine can be reflected as behavioral disinhibition [50–52]. In their elevated plus-maze study, rats chronically treated with nicotine spent a significantly larger percentage of the total arm time on open arms and made significantly more entries to open arms compared with those receiving vehicle. Since these results were expressed as the ratio between open and closed arm entries, the increase in open arm activities was proposed to reflect increased impulsivity

independent of any non-specific changes in motor function [51].

Blondel et al. reported that an acute increase in accuracy disappeared after repeated nicotine administration: instead, an increase in impulsive responding predominated [53]. The present studies differ from these previous findings since improved attentional measures remained. Further, our data with nicotine appear to correlate with clinical findings. Nicotine is long known to improve attention, vigilance and short-term memory in patients upon acute exposure as well as in patients with a smoking history. In addition, it is also well documented that nicotine can cause elevated impulsivity in smokers, and higher levels of impulsivity are associated with continued smoking [54].

4.2. *Ciproxifan studies*

H_3R agonists can impair cognition in various tasks such as object recognition, passive avoidance [55] and social memory [56], whereas H_3R antagonists can rescue impairments produced pharmacologically or genetically. For example, the H_3R antagonists clobenpropit and thioperamide reverse scopolamine-induced deficits in passive avoidance, novel object recognition and elevated plus maze in mice and rats [37,57], as well as deficits in acquisition of a passive avoidance response in senescence accelerated mice [58]. The antagonist, ciproxifan, as well as the Abbott antagonists, A-304121, A-317920 and ABT-239, can improve cognitive performance in the absence of a pharmacological impairment in a short-term social memory task in rats and in a genetic model of ADHD in spontaneously hypertensive rat pups [35,36]. However, it is difficult to definitively dissociate effects on attention from impulsivity in some of these previous studies.

Our 5-CSRTT data with ciproxifan now bring additional insight to the histaminergic H_3R field regarding the effects of blocking H_3Rs on attention and impulsivity. The only previous report examining ciproxifan in the 5-CSRTT was a very limited study with a single dose and a single outcome measure [42]. While that study reported improved attention as measured by increased accuracy (% correct), replication of this finding for ciproxifan or any other H_3R antagonist has not since been reported. In the present studies, we now demonstrate improved attention in experiments conducted at Wyeth Research and decreased impulsivity in experiments conducted at both Abbott and Wyeth. Improved attention was observed when stimulus durations were presented randomly to the rats, thus confirming earlier findings from Ligneau and co-workers [31], albeit under more demanding test conditions. Interestingly, when a non-variable stimulus was used at Abbott, and despite similar baseline responding to rats trained at Wyeth, no improvement in any attentional measure was observed at Abbott. However, of most interest, ciproxifan significantly reduced anticipatory or premature responding without affecting response latencies. This was apparent at Wyeth with a relatively short stimulus duration of 0.2 s and at Abbott in two separate studies with stimulus duration of either 0.5 or 0.25 s. These effects on impulsive behavior are intriguing, have not been reported previously, and indicate that this mechanism may offer additional benefit in ADHD patients compared to other therapies or other novel approaches to treating this disorder.

H_3R antagonists have been proposed as antiobesity agents, so it is possible that potential appetite suppressant effects of ciproxifan may confound outcome measures in the 5-CSRTT, in which food is used as a motivator. Bearing this concern in mind, the physiological effects of ciproxifan have been extensively studied at Abbott [59] in which acute and chronic effects of ciproxifan on food intake, water consumption, body temperature and spontaneous locomotor activity were evaluated in rats. Results showed that an acute high dose of ciproxifan (10 mg/kg but not 3 mg/kg) induced hypoactivity as well as reduced food intake; tolerance to these effects developed following repeated dosing [59]. In the current studies, efficacy (improved attention, decreased impulsivity) was observed at doses of 3 mg/kg and lower, so non-specific effects on food/water intake, body temperature or spontaneous locomotor activity are likely not a confound in our studies.

Histamine H_3Rs are located on histaminergic and non-histaminergic neurons in intermediate and deep layers of the cerebral cortex, striatum, nucleus accumbens, hippocampus and thalamus [60]. H_3 receptors directly modify GABA release from cortical interneurons and antagonism of these H_3 receptors produce decreases in GABA release and disinhibition of the cortical cholinergic system resulting in increased acetylcholine levels [61]. Indeed, histamine application into the medial septum results in a significant reduction in acetylcholine release within the hippocampus that can be blocked with the H_3 antagonist thioperamide, presumably acting via presynaptic autoreceptors [37]. Thus, it is possible that at least some of the observed effects of ciproxifan in the present 5-CSRTT studies may reflect an influence over cholinergic neurotransmission. In support of this, using microdialysis techniques in freely moving rats, we have previously observed increased release of acetylcholine in the prefrontal cortex following administration of H_3R antagonists such as ABT-239 [33], and ciproxifan (unpublished observations); the prefrontal cortex is a brain region key to performance in the 5-CSRTT [9]. Intriguingly, we have also observed increased release of dopamine in the prefrontal cortex, but not the striatum, following administration of ABT-239. This same H_3R antagonist also blocked methamphetamine-induced hyperactivity, further linking behavioral efficacy to the dopaminergic system. Since previous reports also implicate the dopaminergic system in mediating aspects of impulsivity in the 5-CSRTT [12,62], it is possible that blockade of H_3 heteroreceptors play an important role in mediating the decreased impulsive behaviors observed in the present studies with ciproxifan.

In summary, the present studies confirm pro-attentive effects of nicotine in a preclinical assay with relevance for ADHD, the 5-CSRTT. In addition, improved attention and decreased impulsivity were observed with the H_3R antagonist, ciproxifan, in the same test. Since new therapeutic approaches to ADHD and schizophrenia currently in clinical development are targeting nicotinic receptors (e.g. selective $\alpha4\beta2$ neuronal nicotinic receptor agonists, ABT-089 and TC-1734) or histaminergic receptors (e.g. selective histamine H_3R antagonist, GSK-189254), it will be important to determine how preclinical data with these mechanisms translates to humans.

Acknowledgements

The neuronal histamine H_3 receptor (H_3R) has attracted much interest in recent years as a target for new drugs aimed at treating an array of CNS disorders. To a large degree, preclinical evidence for efficacy following blockade of H_3Rs with selective antagonists in animal models of attention deficit hyperactivity disorder (ADHD), schizophrenia, narcolepsy and Alzheimer's disease (AD) has driven this research, which is now leading to early clinical trials with several drug-like compounds. An important influence in this field was Arthur A. Hancock, who led the H_3 team at Abbott Laboratories from the late 1990s to his untimely passing in 2005. Art actively participated in the histaminergic field and encouraged publication and discussion of preclinical efficacy data to help move the H_3 field forward. Indeed, his final review published posthumously in this *Journal* represents a candid account of drug discovery for this target at Abbott. It is in this spirit of communication across laboratories that this article took form as a collaborative effort between scientists at Wyeth Research and Abbott Laboratories. As colleagues of Art's, we know he would appreciate sharing of such knowledge whenever possible.

REFERENCES

[1] Banaschewski T, Brandeis D, Heinrich H, Albrecht B, Brunner E, Rothenberger A. Association of ADHD and conduct disorder—brain electrical evidence for the existence of a distinct subtype. J Child Psychol Psychiatry 2003;44:356–76.
[2] Butler GKL, Montgomery AMJ. Subjective self-control and behavioural impulsivity coexist in anorexia nervosa. Eating Behav 2005;6:221–7.
[3] Egeland J, Rund BR, Sundet K, Landro NI, Asbjornsen A, Lund A, et al. Attention profile in schizophrenia compared with depression: differential effects of processing speed, selective attention and vigilance. Acta Psychiatr Scand 2003;108:276–84.
[4] Retz W, Retz-Junginger P, Hengesch G, Schneider M, Thome J, Pajonk FG, et al. Psychometric and psychopathological characterization of young male prison inmates with and without attention deficit/hyperactivity disorder. Eur Arch Psychiatry Clin Neurosci 2004;V254:201–8.
[5] Sunderland T, Weingartner H, Cohen RM, Tariot PN, Newhouse PA, Thompson KE, et al. Low-dose oral lorazepam administration in Alzheimer subjects and age-matched controls. Psychopharmacology (Berl) 1989;99:129–33.
[6] Villardita C, Grioli S, Lomeo C, Cattaneo C, Parini J. Clinical studies with oxiracetam in patients with dementia of Alzheimer type and multi-infarct dementia of mild to moderate degree. Neuropsychobiology 1992;25:24–8.
[7] Pliszka SR. Psychiatric comorbidities in children with attention deficit hyperactivity disorder: implications for management. Paediatr Drugs 2003;5:741–50.
[8] Green MF, Kern RS, Heaton RK. Longitudinal studies of cognition and functional outcome in schizophrenia: implications for MATRICS. Schizophr Res 2004;72:41–51.
[9] Dervaux A, Bayle FJ, Laqueille X, Bourdel M-C, Le Borgne M-H, Olie J-P, et al. Is substance abuse in schizophrenia related to impulsivity, sensation seeking, or anhedonia? Am J Psychiatry 2001;158:492–4.
[10] Baxter D, Appleby L. Case register study of suicide risk in mental disorders. Br J Psychiatry 1999;175:322–6.
[11] Verdoux H, Liraud F, Gonzales B, Assens F, Abalan F, van Os J. Suicidality and substance misuse in first-admitted subjects with psychotic disorder. Acta Psychiatr Scand 1999;100:389–95.
[12] Robbins TW. The 5-choice serial reaction time task: behavioural pharmacology and functional neurochemistry. Psychopharmacology 2002;163:362–80.
[13] Levinoff EJ, Saumier D, Chertkow H. Focused attention deficits in patients with Alzheimer's disease and mild cognitive impairment. Brain Cogn 2005;57:127–30.
[14] Lee J, Park S. The role of stimulus salience in CPT-AX performance of schizophrenia patients. Schizophr Res 2006;81:191–7.
[15] Mass R. The vigilance paradigm in schizophrenia research—studies on the continuous performance test (CPT). Fortschr Neurol Psychiatr 2002;70:34–9.
[16] Nestor PG, Faux SF, McCarley RW, Shenton ME, Sands SF. Measurement of visual sustained attention in schizophrenia using signal detection analysis and a newly developed computerized CPT task. Schizophr Res 1990;3:329–32.
[17] Nieuwenstein MR, Aleman A, de Haan EH. Relationship between symptom dimensions and neurocognitive functioning in schizophrenia: a meta-analysis of WCST and CPT studies. Wisconsin card sorting test. Continuous performance test. J Psychiatr Res 2001;35:119–25.
[18] Loo SK, Hopfer C, Teale PD, Reite ML. EEG correlates of methylphenidate response in ADHD: association with cognitive and behavioral measures. J Clin Neurophysiol 2004;21:457–64.
[19] Riccio CA, Waldrop JJ, Reynolds CR, Lowe P. Effects of stimulants on the continuous performance test (CPT): implications for CPT use and interpretation. J Neuropsychiatry Clin Neurosci 2001;13:326–35.
[20] Teicher MH, Lowen SB, Polcari A, Foley M, McGreenery CE. Novel strategy for the analysis of CPT data provides new insight into the effects of methylphenidate on attentional states in children with ADHD. J Child Adolesc Psychopharmacol 2004;14:219–32.
[21] Epstein JN, Conners CK, Hervey AS, Tonev ST, Arnold LE, Abikoff HB, et al. Assessing medication effects in the MTA study using neuropsychological outcomes. J Child Psychol Psychiatry 2006;47:446–56.
[22] Bromley E. A collaborative approach to targeted treatment development for schizophrenia: a qualitative evaluation of the NIMH-MATRICS project. Schizophr Bull 2005;31:954–61.
[23] Buchanan RW, Davis M, Goff D, Green MF, Keefe RS, Leon AC, et al. A summary of the FDA-NIMH-MATRICS workshop on clinical trial design for neurocognitive drugs for schizophrenia. Schizophr Bull 2005;31:5–19.
[24] Green MF, Nuechterlein KH. The MATRICS initiative: developing a consensus cognitive battery for clinical trials. Schizophr Res 2004;72:1–3.
[25] Morton WA, Stockton GG. Methylphenidate abuse and psychiatric side effects. Prim Care Companion J Clin Psychiatry 2000;2:159–64.
[26] Stolerman IP, Mirza NR, Hahn B, Shoaib M. Nicotine in an animal model of attention. Eur J Pharmacol 2000;393: 147–54.
[27] Esbenshade TA, Fox GB, Cowart MD. Histamine H3 receptor antagonists: preclinical promise for treating obesity and cognitive disorders. Mol Interv 2006;6:77–88. 59.
[28] Huang ZL, Mochizuki T, Qu WM, Hong ZY, Watanabe T, Urade Y, et al. Altered sleep–wake characteristics and lack of arousal response to H3 receptor antagonist in histamine H1 receptor knockout mice. Proc Natl Acad Sci USA 2006;103:4687–92.

[29] Lamberty Y, Margineanu DG, Dassesse D, Klitgaard H. H3 agonist immepip markedly reduces cortical histamine release, but only weakly promotes sleep in the rat. Pharmacol Res 2003;48:193–8.
[30] Leurs R, Blandina P, Tedford C, Timmerman H. Therapeutic potential of histamine H3 receptor agonists and antagonists. Trends Pharmacol Sci 1998;19:177–83.
[31] Lin JS, Sakai K, Vanni-Mercier G, Arrang JM, Garbarg M, Schwartz JC, et al. Involvement of histaminergic neurons in arousal mechanisms demonstrated with H3-receptor ligands in the cat. Brain Res 1990;523:325–30.
[32] Passani MB, Lin JS, Hancock A, Crochet S, Blandina P. The histamine H3 receptor as a novel therapeutic target for cognitive and sleep disorders. Trends Pharmacol Sci 2004;25:618–25.
[33] Fox GB, Esbenshade TA, Pan JB, Radek RJ, Krueger KM, Yao BB, et al. Pharmacological properties of ABT-239 [4-(2-{2-[(2R)-2-methylpyrrolidinyl]ethyl}-benzofuran-5-yl)benzonitrile]. II. Neurophysiological characterization and broad preclinical efficacy in cognition and schizophrenia of a potent and selective histamine H3 receptor antagonist. J Pharmacol Exp Ther 2005;313:176–90.
[34] Schwartz JC, Morisset S, Rouleau A, Tardivel-Lacombe J, Gbahou F, Ligneau X, et al. Application of genomics to drug design: the example of the histamine H3 receptor. Eur Neuropsychopharmacol 2001;11:441–8.
[35] Fox GB, Pan JB, Faghih R, Esbenshade TA, Lewis A, Bitner RS, et al. Identification of novel H3 receptor (H3R) antagonists with cognition enhancing properties in rats. Inflamm Res 2003;52(Suppl 1):S31–2.
[36] Fox GB, Pan JB, Radek RJ, Lewis AM, Bitner RS, Esbenshade TA, et al. Two novel and selective nonimidazole H3 receptor antagonists A-304121 and A-317920. II. In vivo behavioral and neurophysiological characterization. J Pharmacol Exp Thera 2003;305:897–908.
[37] Giovannini MG, Bartolini L, Bacciottini L, Greco L, Blandina P. Effects of histamine H3 receptor agonists and antagonists on cognitive performance and scopolamine-induced amnesia. Behav Brain Res 1999;104:147–55.
[38] Hancock AA, Fox GB. Perspectives on cognitive domains, H3 receptor ligands and neurological disease. Expert Opin Investig Drugs 2004;13:1237–48.
[39] Komater VA, Buckley MJ, Browman KE, Pan JB, Hancock AA, Decker MW, et al. Effects of histamine H3 receptor antagonists in two models of spatial learning. Behav Brain Res 2005;159:295–300.
[40] Orsetti M, Ferretti C, Gamalero R, Ghi P. Histamine H3-receptor blockade in the rat nucleus basalis magnocellularis improves place recognition memory. Psychopharmacology (Berl) 2002;159:133–7.
[41] Passani MB, Blandina P. Cognitive implications for H3 and 5-HT3 receptor modulation of cortical cholinergic function: a parallel story. Meth Find Exp Clin Pharmacol 1998;20:725–33.
[42] Ligneau X, Lin J, Vanni-Mercier G, Jouvet M, Muir JL, Ganellin CR, et al. Neurochemical and behavioral effects of ciproxifan, a potent histamine H3-receptor antagonist. J Pharmacol Exp Ther 1998;287:658–66.
[43] Kirkby DL, Jones DN, Barnes JC, Higgins GA. Effects of anticholinesterase drugs tacrine and E2020, the 5-HT(3) antagonist ondansetron, and the H(3) antagonist thioperamide, in models of cognition and cholinergic function. Behav Pharmacol 1996;7:513–25.
[44] Hahn B, Shoaib M, Stolerman IP. Nicotine-induced enhancement of attention in the five-choice serial reaction time task: the influence of task demands. Psychopharmacology (Berl) 2002;162:129–37.
[45] Mirza NR, Ian PS. Nicotine enhances sustained attention in the rat under specific task conditions. Psychopharmacology 1998;V138:266–74.
[46] Shoaib M, Bizarro L. Deficits in a sustained attention task following nicotine withdrawal in rats. Psychopharmacology (Berl) 2005;178:211–22.
[47] Young JW, Finlayson K, Spratt C, Marston HM, Crawford N, Kelly JS, et al. Nicotine improves sustained attention in mice: evidence for involvement of the alpha7 nicotinic acetylcholine receptor. Neuropsychopharmacology 2004;29:891–900.
[48] McCann MF, Irwin DE, Walton LA, Hulka BS, Morton JL, Axelrad CM. Nicotine and cotinine in the cervical mucus of smokers, passive smokers, and nonsmokers. Cancer Epidemiol Biomarkers Prev 1992;1:125–9.
[49] Thompson JG, Irwin FD, Kanematsu S, Seraydarian K, Suh M. Effects of chronic nicotine administration and age in male Fischer-344 rats. Toxicol Appl Pharmacol 1973;26:606–20.
[50] Ericson M, Olausson P, Engel JA, Soderpalm B. Nicotine induces disinhibitory behavior in the rat after subchronic peripheral nicotinic acetylcholine receptor blockade. Eur J Pharmacol 2000;397:103–11.
[51] Olausson P, Engel JA, Soderpalm B. Behavioral sensitization to nicotine is associated with behavioral disinhibition; counteraction by citalopram. Psychopharmacology (Berl) 1999;142:111–9.
[52] Olausson P, Ericson M, Lof E, Engel JA, Soderpalm B. Nicotine-induced behavioral disinhibition and ethanol preference correlate after repeated nicotine treatment. Eur J Pharmacol 2001;417:117–23.
[53] Blondel A, Sanger DJ, Moser PC. Characterisation of the effects of nicotine in the five-choice serial reaction time task in rats: antagonist studies. Psychopharmacology (Berl) 2000;149:293–305.
[54] Mitchell SH. Effects of short-term nicotine deprivation on decision-making: delay, uncertainty and effort discounting. Nicotine Tob Res 2004;6:819–28.
[55] Blandina P, Giorgetti M, Bartolini L, Cecchi M, Timmerman H, Leurs R, et al. Inhibition of cortical acetylcholine release and cognitive performance by histamine H3 receptor activation in rats. Br J Pharmacol 1996;119:1656–64.
[56] Prast H, Argyriou A, Philippu A. Histaminergic neurons facilitate social memory in rats. Brain Res 1996;734:316–8.
[57] Miyazaki S, Imaizumi M, Onodera K. Effects of thioperamide, a histamine H3-receptor antagonist, on a scopolamine-induced learning deficit using an elevated plus-maze test in mice. Life Sci 1995;57:2137–44.
[58] Meguro K, Yanai K, Sakai N, Sakurai E, Maeyama K, Sasaki H, et al. Effects of thioperamide, a histamine H3 antagonist, on the step-through passive avoidance response and histidine decarboxylase activity in senescence-accelerated mice. Pharmacol Biochem Behav 1995;50:321–5.
[59] Pan JB, Yao BB, Miller TR, Kroeger PE, Bennani YL, Komater VA, et al. Evidence for tolerance following repeated dosing in rats with ciproxifan, but not with A-304121. Life Sci 2006;79:1366–79.
[60] Pillot C, Ortiz J, Heron A, Ridray S, Schwartz JC, Arrang JM. Ciproxifan, a histamine H3-receptor antagonist/inverse agonist, potentiates neurochemical and behavioral effects of haloperidol in the rat. J Neurosci 2002;22:7272–80.
[61] Giorgetti M, Bacciottini L, Bianchi L, Giovannini MG, Cecchi M, Blandina P. GABAergic mechanism in histamine H3 receptor inhibition of K(+)-evoked release of acetylcholine from rat cortex in vivo. Inflamm Res 1997;46(suppl 1):S33–4.
[62] Cole BJ, Robbins TW. Effects of 6-hydroxydopamine lesions of the nucleus accumbens septi on performance of a 5-choice serial reaction time task in rats: implications for theories of selective attention and arousal. Behav Brain Res 1989;33:165–79.