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Learning and Memory Performance Following Acute Intranasal Insulin Administration in Abstinent Smokers

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Abstract

The highest incidence of relapse to smoking occurs within the first 2 weeks of a cessation attempt. In addition to enhanced nicotine craving, this phase of smoking cessation is also marked by learning and memory dysfunction. Many smokers are not able to overcome these symptoms, and they relapse to smoking shortly after trying to quit. In two clinical studies, we evaluated intranasal insulin for efficacy in improving learning and memory function during nicotine withdrawal. Our first study was a crossover evaluation (N=19) following 20 hours of smoking abstinence. Study 2 was a parallel design study (N=50) following 16 hours of abstinence. Both studies were randomized and placebo-controlled, in which the 60 IU intranasal insulin dose was administered and cognitive function was measured using California Verbal Learning Test-II. Intranasal insulin did not improve learning over the five verbal learning trials. In addition, intranasal insulin did not improve either short- or long-delay recall in either study. In summary, the one-time administration of intranasal insulin does not improve **verbal** learning and memory in smokers. Whether longer administration schedules may be of benefit should be evaluated in future studies.

INTRODUCTION

Many cigarette smokers express a desire to quit smoking, however rates of relapse to smoking within 1 year after initiating a quit attempt are ~90% (Hughes, Goldstein, Hurt, Shiffman, 1999), with the highest incidence of relapse occurring within the first 2 weeks (Swan, Ward, Carmelli and Jack, 1993). A significant barrier to quitting for many smokers is the tobacco abstinence syndrome, marked by nicotine craving, anxiety, restlessness, and cognitive impairment (Myers et al., 2008; Merritt et al., 2010; Hirshman et al., 2004; Ernst et al., 2001). One important dysfunction, which we studied here, is an impairment in verbal learning and memory during nicotine withdrawal.

Verbal memory function is assessed by a task that involves presentation of a list of words that a subject is asked to recall immediately as well as after a delay. Previous studies have shown that abstinent smokers are impaired in recalling presented words. Evaluating a cohort of adolescent smokers under conditions of *ad lib* smoking and after 24 hours of abstinence,

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Jacobsen et al (2007) have shown a decline in immediate verbal memory under the condition of smoking abstinence. In an additional study involving *ad lib* smokers, abstinent smokers and nonsmokers who completed a verbal learning protocol, Soar et al (2008) have shown that the impairment in verbal learning associated with smoking abstinence is reversed following smoking resumption. These studies utilized Hopkins Verbal Learning Test – Revised (Jacobsen et al., 2008) and Auditory Verbal Learning Test (Soar et al., 2008), in which, similarly to the test utilized in the present study, a participant first learns a list of words (following the list being presented five times), and then recalls it immediately or after a period of delay (i.e. less than 30 minutes).

Nicotine improves memory function by binding to presynaptic nicotinic acetylcholine receptors in the brain, thereby facilitating the release of acetylcholine, dopamine, serotonin, glutamate, and other neurotransmitters known to be involved in cognitive processes (Di Matteo et al., 2007). A meta-analysis, conducted by Heishman et al., (2010) has shown that the effect of nicotine on verbal memory is due to its true cognition-enhancing effect, not as part of a more general withdrawal relief. The meta-analysis only included studies of nonsmokers and smokers who were either not tobacco-deprived or deprived for less than 2 hours; thereby removing the confound of nicotine withdrawal.

In this investigation, we have focused on intranasal insulin as part of studying a novel treatment which would replace nicotine's pro-cognitive effects during smoking cessation. In fact, the finding that intranasal insulin clinically improves verbal memory function has been replicated. In both studies (Craft et al., 2012; Craft et al., 2017), patients with Alzheimer's disease dementia or amnesic mild cognitive impairment had better composite verbal memory scores following chronic intranasal insulin treatment in comparison to the placebo-treated group. It is hypothesized that such effect is observed due to insulin's effect on preventing neuro-inflammatory and neuro-toxic molecular mechanisms as shown in the rodent model of Alzheimer's disease (Rajasekar et al., 2016).

Based on the earlier findings that verbal memory is impaired during abstinence from smoking, and that intranasal insulin improves verbal memory in clinical populations, we here investigated intranasal insulin during acute abstinence from smoking, hypothesizing that intranasal insulin would improve learning and memory functions during withdrawal.

Insulin can be effectively delivered directly to the brain via the intranasal route, which permits the peptide to reach the brain via the olfactory nerve pathways leading from the nasal cavity directly to the central nervous system (Dhuria et al., 2009) with insignificant systemic absorption and hypoglycemic events. Cerebrospinal fluid insulin in humans peaks 30 minutes following the intranasal administration without peripheral insulin changes (Born et al., 2002). In our primary work, we have shown that intranasal insulin relieves nicotine cravings (Hamidovic et al., 2017). We here report the results of our investigation on intranasal insulin efficacy in improving verbal learning and memory function during nicotine withdrawal.

METHODS

Participants

Male and female smokers living in Central New Mexico (18 to 65 years of age) were recruited via advertisement to participate. Subjects were included if they reported smoking 10 cigarettes per day for the past year, had normal vitals (blood pressure <140/90, heart rate 50-100 bpm, body temperature <37.4C), normal blood glucose (80-140 mg/dL), normal or overweight body mass index (18.5-30 kg/m²) and a CO level > 6ppm. Subjects were excluded for previous/current insulin use, Diagnostic Statistical Manual-IV-Revised Axis I disorder or high suicidal ideation, current pregnancy or lactation, Michigan Alcoholism Screening Test (MAST) score >5, current use of illicit drugs (verified using a urine sample), current use of a smoking cessation aid (nicotine replacement therapy, Chantix or Wellbutrin), current prescription or over-the-counter medication use, nasal pathology, hyposmia or anosmia (score <10 on Sniffin' Sticks test of olfactory function), Shipley-2 standard score of less than 80, Blood alcohol concentration >0.00% (using AlcoSensor FST Breathalyzer), lifetime history of endocrine disease, or concomitant allergies. Additionally, Study 1 subjects were excluded for eating disorders, as well as if they had Three-Factor Eating Questionnaire disinhibition score >8 or restraint score >11. The FDA approved the Investigational New Drug application (IND 120700 and IND 116626-Study 2). University of New Mexico Human Research Protections Office also approved the study. All study participants in both studies signed the consent form prior to study participation.

Study 1 Procedures

Subjects first attended a screening session to ensure study eligibility. Following the screening appointment, subjects were admitted to an inpatient unit for 1 overnight stay. Subjects reported to the inpatient unit of University of New Mexico Clinical and Translational Sciences Center at 1130 h on day 1 after beginning smoking cessation at 2000 h on the day prior. Following confirmation of overnight smoking abstinence with the CO level being 50% of the level detected at the screening session, subjects provided urine and breath samples to rule out illicit drug use and alcohol consumption. In order to continue with the study session, study participants had to have vitals within normal limits (BP < 140/90 mmhg, HR between 50-100 bpm, and temperature <37.5 C). In addition, as agreed upon with the FDA, study participants had to have a normal olfactory physical exam and a point-of-care glucose level > 70 mg/dL in order to proceed with treatment administration. Spray was administered at 1240 h as one spray in each nostril every three minutes for the total of six sprays. Lunch was given at 1300 h and CVLT-II was administered at 1320 h. The timing of CVLT-II administration was projected to coincide with the peak CSF insulin level following intranasal administration (Born et al., 2002). Discharge occurred at 1245 h on day 2 following administration of additional study tests (published in Hamidovic et al., 2017). The entire session was 36 h of abstinence; the CVLT-II testing occurred approximately after 20 hours of abstinence. The two sessions (placebo and insulin, in random order) of this crossover study were separated by approximately 1 week.

Study 2 Procedures

This paper is an analysis of a two-session study evaluating efficacy of intranasal insulin (60 IU). The first session involved an evaluation of learning and memory (details outlined below). The second visit involved participation in a psychosocial stress session. The measures from that session - craving, hormonal and cardiovascular responses - are published separately in Hamidovic et al (2017).

Study subjects attended a screening appointment and an outpatient study session. The screening session included two parts. First, eligibility criteria – as specified in the “Participants” section – was assessed. Upon completion of the 3 ½ h screening visit, subjects were scheduled for the first of the total of two study sessions. The procedures and results presented here are from the first study session. Participants (n=50) reported to the outpatient unit of the University of New Mexico Clinical and Translational Sciences Center at 0800 h after beginning smoking abstinence at 2000 h the night prior. Abstinence from smoking was verified with a CO reading on a coVita (Haddonfield, NJ, USA) piCO+ breathalyzer with less than 50% of their baseline CO recorded at intake. A subset of individuals (N =24 of the sample) had a nicotine level verified by a serum analysis. None of the tested subjects had a detectable nicotine level suggestive of smoking. Following negative urine drug and pregnancy tests, at 0900 h, subjects were then provided breakfast (a slice of banana bread, 1 apple and 2 cheese sticks) that they had to eat entirely. The purpose of providing breakfast was to ensure food intake as study participants underwent a fast until the session was completed. The rest of the morning, subjects watched and read emotionally neutral movies and literature.

At 1215 h, a study nurse collected the first blood sample and completed a POC glucose check and nares assessment. As agreed upon with the FDA, the purpose of the POC glucose check was to ensure that none of the tested participants were hypoglycemic prior to intranasal insulin administration. At the time of the study, it was not known whether the treatment may cause a drop in blood glucose. In addition, the FDA required that a nares check be performed prior to intranasal insulin administration to rule out inflammation or infection.

Intranasal spray administration of insulin or placebo was carried out at 1230 h as one spray in each nostril every 3 minutes for the total of 6 sprays. At 1310 h, CVLT-II was administered at the time projected to reflect peak CSF insulin levels (Born et al., 2002). The second blood sample was collected at 1420 h. Study participants were discharged at 1600 h with two POC measurements and a snack between 1420 h and 1600 h. As agreed upon with the FDA, subjects were kept in the lab even though data collection ended to ensure that they would not be sent home in case of a sudden hypoglycemic episode.

Drug Dosing and Preparation

Drug compounding occurred at the University of New Mexico Health Sciences Center Translational Pharmacy lab in a biosafety cabinet using aseptic technique per FDA-approved protocol. The drug substance used for Intranasal Insulin was bulk Novolin R (Novo Nordisk, Plainsboro, NJ, USA). Drug preparation occurred within 30 minutes of administration. The

drug was packaged in 10 ml amber glass bottles (Gerresheimer, Dusseldorf, Germany) capped with 10 mg nasal pumps (Aero Pump, Hochheim am Main, Germany). The dose was selected based on a study in Alzheimer's disease patients in which the doses were 20 and 40 IU (Craft et al., 2012). Though higher doses (160 IU) have been used (Benedict et al., 2004; Benedict et al., 2008), those doses have been associated with increased circulating insulin immediately following intranasal administration. We avoided administering the high dose of intranasal insulin out of the concern of hypoglycemia. A full description of drug preparation and administration may be found in our original publication (Hamidovic et al., 2017).

California Verbal Learning Test-II Adult Version

Learning and memory were assessed using the California Verbal Learning Test-II Adult version (CVLT-II) (Delis, Kramer, Kaplan & Ober, 2000). The CVLT-II is a neuropsychological test of episodic memory, which involves the oral presentation of a 16-word list (List A) over five immediate-recall trials (Trials 1-5). An interference list (List B) is then presented for one immediate-recall trial (Trial B), followed by free-recall (participant asked to recite as many words as they could remember in any order) of List A (Short Delay Free Recall), and a cued recall (participant prompted to recite words from four semantic categories) of List A (Short Delay Cued Recall). After a 20-minute delay, subjects are asked to free-recall list A (Long Delay Free Recall), followed by cued-recall of List A (Long Delay Cued Recall). (See Figure 1).

Test administration time was about 45 minutes, including the 20-minute delay interval during which subjects completed questionnaires. Audio recordings for each CVLT-II were used for scoring after the test was completed. Two trained research coordinators listened to the recordings independently and then compared results for accuracy. Discrepancies were re-listened by the research coordinators following which the results were synchronized. Scores were reported for trial 1-5, trial B, short delay free recall, short delay cued recall, long delay free recall and long delay cued recall.

Data Analysis

Participant characteristics were compared using T-test for continuous and chi-square for categorical variables. All outcome data were tested for distribution normality and skewness. If detected, data were corrected using the *lnskew0* STATA command, which performs a log transformation after adding a constant, thus creating a zero skewness logged variable. Before continuing with data analysis, it was first confirmed that this method did indeed correct variable normality and skewness.

Study 1 data analyses were carried out using mixed model analyses. Random coefficient models were constructed for analyzing study outcomes as a combination of fixed and random effects. Learning scores included the number of recalled words (over five trials). Learning was analyzed in a model with treatment, order of treatment administration, trial, and treatment by trial interaction as fixed effects. Trial nested within subject and trial nested within treatment nested within subject were included as random effects. Trial B, short and long delay (both free and cued) analysis models included treatment and baseline scores as fixed effects, and treatment nested within subject as random effect.

Study 2 data analysis was carried out using mixed model analyses. Learning was analyzed in a model with treatment, trial, and treatment by trial interaction as fixed effects. Trial nested within subject was included as random effect. Trial B, short and long delay (both free and cued) analysis models included treatment and baseline scores as fixed effect. Treatment nested within subject was included as a random term. Sample size was determined a priori using Soar et al (2008). The result of that analysis showed an approximate size of 25 participants per group to capture the effect of treatment. One subject's CVLT-II data in Study 2 was discarded because the subject had zero words recalled, suggestive of unwillingness to participate in the study procedure.

Data were analyzed using STATA Version 12. The p statistics are reported at their nominal, non-multiple comparison- corrected value.

RESULTS

Subjects

Study 1 basic demographic table is published in Hamidovic et al (2017) and summarized in Table 1 of that manuscript.

Twenty-six of the total of fifty subjects completing Study 2 received intranasal insulin (see Table 1). Most participants were White men of middle age who were moderately nicotine dependent. T-test and chi-square analyses for continuous and categorical variables (respectively) listed in Table 1 did not reveal any significant group differences. In addition to basic demographic variables, Table 1 also shows lifetime use of illicit drugs by treatment groups.

Learning

Intranasal insulin did not improve learning in either study as evidenced by a lack of either main effect of treatment or a treatment $\times$ trial interaction. As expected, study participants learned new words upon the list being repeated over 5 trials. The graphical details are displayed in Figures 2 and 3 (left side) for studies 1 and 2, respectively. The details of analytical results are displayed in Tables 2 and 4 for studies 1 and 2, respectively.

Memory

Intranasal insulin did not improve short- and long-term recall in either study. The graphical details are displayed in Figures 2 and 3 (right side) for studies 1 and 2, respectively. The details of analytical results are displayed in Tables 3 and 5 for studies 1 and 2, respectively. The non-significant effect of treatment was the case for both free and cued recall.

DISCUSSION

In summary, intranasal insulin administration did not improve verbal learning or memory in our studies designed based on rigorous clinical trial methodology. Our hypothesis that intranasal insulin would improve these outcomes was based on earlier studies in Alzheimer's disease dementia or amnesic mild moderate impairment patients showing memory improvements following intranasal insulin treatment (Craft et al., 2012; Craft et al., 2017).

It could be argued that we did not observe significant effects because they may not be observable after an acute, one-time intranasal insulin administration. This is certainly a possibility since the literature regarding memory task performance following one-time intranasal insulin administration is mixed. For example, Reger et al (2006) found that one intranasal insulin dose (20 IU) caused worse word recall scores, while the 40 IU dose caused an improvement in word recall. Moreover, whereas Brunner et al (2015) were able to show that intranasal insulin (40 IU), administered to healthy young men, improved delayed (but not immediate) odor-cued recall of spatial memory, the group (Brunner et al (2016)) was not able to replicate the behavioral data (recalling olfactory or visual cues at different target locations) in a functional brain imaging study.

A recent study (Feld et al., 2016) evaluated whether sleep and intranasal insulin (160 IU) enhance memory consolidation. Just prior to going to sleep, study participants learned word pairs and were given intranasal insulin. The following evening, they learned new pairs. Whereas insulin did not enhance original word pair recall, it impaired the acquisition of new pairs. The authors concluded that insulin acts on mechanisms that diminish the subsequent encoding of novel information. Whereas the overall lack of insulin's efficacy on recall matches what we found in our study, the interference result is different from what we observed. In the CVLT-II protocol, List B – the interference list – was not found to be different between the insulin and the placebo condition. One possibility is that sleep (implemented in the Feld et al., 2016 study) is a requirement to observe the action of insulin in disrupting interfering information. The other possibility is that the dose we used in our study is inadequate to observe this specific effect.

Efficacy of *chronic* intranasal insulin seems to be not only disease-specific, but may also reflect medication adherence. Though the study that intranasal insulin improved memory function in Alzheimer's disease dementia or amnesic mild moderate impairment has been replicated (Craft et al., 2012; Reger et al., 2008), the negative findings of intranasal insulin on other clinical outcomes, including Bipolar Disorder (McIntyre et al., 2012), Major Depressive Disorder (Cha et al., 2017), Schizophrenia (Fan et al., 2013) need to be evaluated in light of a substantial concern regarding medication adherence in the clinical trials with intranasal insulin. Currently, intranasal insulin is formulated with the commercially available insulin designed for subcutaneous administration. In our studies (Hamidovic et al., 2017), we have shown that intranasal insulin causes substantial pain and burning, and others (Lehrer 2015) have called for reformulation of intranasal insulin. Medication adherence is a problem for all clinical trials, but the degree of its interference in the outcome of trials with intranasal insulin needs to be carefully evaluated.

The main limitation of our study is a lack of non-abstinence condition. It is not known whether participants our study experienced a decline in verbal learning and memory. However, numerous studies have shown that abstinent smokers are impaired in recalling presented words immediately as well as after a delay (Wesnes et al., 2013; Soar et al., 2008, Merritt et al 2010; Merritt et al 2012; Dunbar et al., 2007; Foulds et al. 1996; Krebs et al. 1994). Though our study was a single-blind in design, participants were not aware of the treatment. Our placebo was compounded as 8.7% sodium chloride, which produced similar

level of pain and burning (Hamidovic et al., 2017), thereby masking participants' awareness of treatment received.

In conclusion, our rigorous clinical trial methodology studies showed no effect of intranasal insulin on learning and memory functions. These studies involved acute administration of intranasal insulin, which may be different from central insulin's effects observed following a long term administration.

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References

- Benedict C, Hallschmid M, Hatke A, Schultes B, Fehm HL, Born J, Kern W. Intranasal insulin improves memory in humans. *Psychoneuroendocrinology*. 2004; 29(10):1326–1334. DOI: 10.1016/j.psyneuen.2004.04.003 [PubMed: 15288712]
- Benedict C, Kern W, Schultes B, Born J, Hallschmid M. Differential sensitivity of men and women to anorexic and memory-improving effects of intranasal insulin. *Journal of Clinical Endocrinology & Metabolism*. 2008; 93(4):1339–1344. DOI: 10.1210/jc.2007-2606 [PubMed: 18230654]
- Born J, Lange T, Kern W, McGregor GP, Bickel U, Fehm HL. Sniffing neuropeptides: a transnasal approach to the human brain. *Nature Neuroscience*. 2002; 5(6):514–516. DOI: 10.1038/nn849 [PubMed: 11992114]
- Brunner YF, Kofoet A, Benedict C, Freiherr J. Central Insulin Administration Improves Odor-Cued Reactivation of Spatial Memory in Young Men. *Journal of Clinical Endocrinology & Metabolism*. 2015; 100(1):212–219. DOI: 10.1210/jc.2014-3018 [PubMed: 25337926]
- Brunner YF, Rodriguez-Raecke R, Mutic S, Benedict C, Freiherr J. Neural correlates of olfactory and visual memory performance in 3D-simulated mazes after intranasal insulin application. *Neurobiology of Learning and Memory*. 2016; 134:256–263. DOI: 10.1016/j.nlm.2016.07.027 [PubMed: 27492601]
- Cha DS, Best MW, Bowie CR, Gallagher LA, Woldeyohannes HO, Soczynska JK, McIntyre RS. A randomized, double-blind, placebo-controlled, crossover trial evaluating the effect of intranasal insulin on cognition and mood in individuals with treatment-resistant major depressive disorder. *Journal of Affective Disorders*. 2017; 210:57–65. DOI: 10.1016/j.jad.2016.12.006 [PubMed: 28013123]
- Craft S, Baker LD, Montine TJ, Minoshima S, Watson GS, Claxton A, Gerton B. Intranasal Insulin Therapy for Alzheimer Disease and Amnesic Mild Cognitive Impairment A Pilot Clinical Trial. *Archives of Neurology*. 2012; 69(1):29–38. DOI: 10.1001/archneurol.2011.233 [PubMed: 21911655]
- Craft S, Claxton A, Baker LD, Hanson AJ, Cholerton B, Trittschuh EH, Dahl D, Caulder E, Neth B, Montine TJ, et al. Effects of Regular and Long-Acting Insulin on Cognition and Alzheimer's Disease Biomarkers: A Pilot Clinical Trial. *J Alzheimers Dis*. 2017 Mar 29. 2017.
- Delis, C., Kramer, JH., Kaplan, E., Ober, BA. California Verbal Learning Test – second edition Adult version. Psychological Corporation; San Antonio, TX: 2000.
- Dhuria SV, Hanson LR, Frey WH. Novel Vasoconstrictor Formulation to Enhance Intranasal Targeting of Neuropeptide Therapeutics to the Central Nervous System. *Journal of Pharmacology and Experimental Therapeutics*. 2009; 328(1):312–320. DOI: 10.1124/jpet.108.145565 [PubMed: 18945930]
- Dunbar G, Boeijinga PH, Demazieres A, Cisterni C, Kuchibhatla R, Wesnes K, Luthringer R. Effects of TC-1734 (AZD3480), a selective neuronal nicotinic receptor agonist, on cognitive performance and the EEG of young healthy male volunteers. *Psychopharmacology*. 2007; 191(4):919–929. DOI: 10.1007/s00213-006-0675-x [PubMed: 17225162]

- Ernst M, Heishman SJ, Spurgeon L, London ED. Smoking history and nicotine effects on cognitive performance. *Neuropsychopharmacology*. 2001; 25(3):313–319. DOI: 10.1016/s0893-133x(01)00257-3 [PubMed: 11522460]
- Fan XD, Liu E, Freudenreich O, Copeland P, Hayden D, Ghebremichael M, Henderson DC. No Effect of Adjunctive, Repeated-Dose Intranasal Insulin Treatment on Psychopathology and Cognition in Patients With Schizophrenia. *Journal of Clinical Psychopharmacology*. 2013; 33(2):226–230. DOI: 10.1097/JCP.0b013e31828701d0 [PubMed: 23422397]
- Feld GB, Wilhem I, Benedict C, Rudel B, Klameth C, Born J, Hallschmid M. Central Nervous Insulin Signaling in Sleep-Associated Memory Formation and Neuroendocrine Regulation. *Neuropsychopharmacology*. 2016; 41(6):1540–1550. DOI: 10.1038/npp.2015.312 [PubMed: 26448203]
- Foulds J, Stapleton J, Swettenham J, Bell N, McSorley K, Russell MAH. Cognitive performance effects of subcutaneous nicotine in smokers and never-smokers. *Psychopharmacology*. 1996; 127(1):31–38. DOI: 10.1007/bf02805972 [PubMed: 8880941]
- Hamidovic A, Khafaja M, Brandon V, Anderson J, Ray G, Allan AM, Burge MR. Reduction of smoking urges with intranasal insulin: a randomized, crossover, placebo-controlled clinical trial. *Molecular Psychiatry*. 2017; 00:1–9.
- Heishman SJ, Kleykamp BA, Singleton EG. Meta-analysis of the acute effects of nicotine and smoking on human performance. *Psychopharmacology*. 2010; 210(4):453–469. DOI: 10.1007/s00213-010-1848-1 [PubMed: 20414766]
- Hirshman E, Rhodes DK, Zinser M, Merritt P. The effect of tobacco abstinence on recognition memory, digit span recall, and attentional vigilance. *Experimental and Clinical Psychopharmacology*. 2004; 12(1):76–83. DOI: 10.1037/1064-1297.12.1.76 [PubMed: 14769102]
- Jacobsen LK, Mencl WE, Constable RT, Westerveld M, Pugh KR. Impact of smoking abstinence on working memory neurocircuitry in adolescent daily tobacco smokers. *Psychopharmacology*. 2007; 193:557–566. [PubMed: 17505817]
- Krebs SJ, Petros TV, Beckwith BE. EFFECTS OF SMOKING ON MEMORY FOR PROSE PASSAGES. *Physiology & Behavior*. 1994; 56(4):723–727. DOI: 10.1016/0031-9384(94)90233-x [PubMed: 7800739]
- Lehrer, S. Adverse side effects of intranasal detemir insulin in the SNIFF Trial. *J Alzheimers Dis*. 2015. Available at: <http://www.j-alz.com/content/adverse-sideeffects-intranasal-detemir-insulin-sniff-trial>
- McIntyre RS, Soczynska JK, Woldeyohannes HO, Miranda A, Vaccarino A, MacQueen G, Kennedy SH. A randomized, double-blind, controlled trial evaluating the effect of intranasal insulin on neurocognitive function in euthymic patients with bipolar disorder. *Bipolar Disorders*. 2012; 14(7):697–706. DOI: 10.1111/bdi.12006 [PubMed: 23107220]
- Merritt PS, Cobb AR, Cook GI. Sex Differences in the Cognitive Effects of Tobacco Abstinence: A Pilot Study. *Experimental and Clinical Psychopharmacology*. 2012; 20(4):258–263. DOI: 10.1037/a0027414 [PubMed: 22369669]
- Merritt PS, Cobb AR, Moissinac L, Hirshman E. Evidence That Episodic Memory Impairment During Tobacco Abstinence Is Independent of Attentional Mechanisms. *Journal of General Psychology*. 2010; 137(4):331–342. DOI: 10.1080/00221309.2010.499395 [PubMed: 21086856]
- Myers CS, Taylor RC, Moolchan ET, Heishman SJ. Dose-related enhancement of mood and cognition in smokers administered nicotine nasal spray. *Neuropsychopharmacology*. 2008; 33(3):588–598. DOI: 10.1038/sj.npp.1301425 [PubMed: 17443125]
- Hughes, Goldstein, Hurt, Shiffman. in *Recent advances in the pharmacotherapy of smoking* (vol 281, pg 72, 1999). *Jama-Journal of the American Medical Association*. 1999; 281(12):1086–1086.
- Rajasekar N, Nath C, Hanif K, Shukla R. Intranasal Insulin Administration Ameliorates Streptozotocin (ICV)-Induced Insulin Receptor Dysfunction, Neuroinflammation, Amyloidogenesis, and Memory Impairment in Rats. *Mol Neurobiol*. 2016
- Reger MA, Watson GS, Frey WH, Baker LD, Cholerton B, Keeling ML, Craft S. Effects of intranasal insulin on cognition in memory-impaired older adults: Modulation by APOE genotype. *Neurobiology of Aging*. 2006; 27(3):451–458. DOI: 10.1016/j.neurobiolaging.2005.03.016 [PubMed: 15964100]

- Reger MA, Watson GS, Green PS, Wilkinson CW, Baker LD, Cholerton B, Craft S. Intranasal insulin improves cognition and modulates beta-amyloid in early AD. *Neurology*. 2008; 70(6):440–448. DOI: 10.1212/01.wnl.0000265401.62434.36 [PubMed: 17942819]
- Soar K, Dawkins L, Begum H, Parrott AC. The effects of cigarette smoking and abstinence on auditory verbal learning. *Human Psychopharmacology-Clinical and Experimental*. 2008; 23(7):621–627. DOI: 10.1002/hup.968 [PubMed: 18712701]
- Swan GE, Ward MM, Carmelli D, Jack LM. DIFFERENTIAL RATES OF RELAPSE IN SUBGROUPS OF MALE AND FEMALE SMOKERS. *Journal of Clinical Epidemiology*. 1993; 46(9):1041–1053. DOI: 10.1016/0895-4356(93)90172-w [PubMed: 8263577]
- Wesnes KA, Edgar CJ, Kezic I, Salih HM, de Boer P. Effects of nicotine withdrawal on cognition in a clinical trial setting. *Psychopharmacology*. 2013; 229(1):133–140. DOI: 10.1007/s00213-013-3089-6 [PubMed: 23591603]
- Lehrer, S. Adverse side effects of intranasal detemir insulin in the SNIFF Trial; *J Alzheimers Dis*. p. 2015 Available at: <http://www.j-alz.com/content/adverse-sideeffects-intranasal-detemir-insulin-sniff-trial>

Figure 1.

Graphic Presentation of California Verbal Learning Test-II. List A was read to study participants five times. After each list A reading, participants recalled back the words they memorized. List B was a different (i.e. the “distractor” list) read to participants, which they were asked to repeat. Short delay recall refers to recalling list A words immediately after list B was read to the participants. Long delay refers to recalling list A words after a 20-minute break. “Free” recall refers to recalling words without being asked to recall them in a particular categorical order; “cued” recall involves the examiner specifying a particular word category (for example, “animals”), which served as a cue to subjects.

Figure 2.

Graphic Presentation of CVLT-II Performance in Study 1. The study (N=19) was a crossover evaluation of intranasal insulin (60 IU) vs placebo. Participants were tested **after 20 hours** of smoking abstinence. Intranasal insulin had no effect on learning (trial 1-5), trial B (distractor trial), or any of the 4 recall trials as listed in the figure.

Figure 3.

Graphic Presentation of CVLT-II Performance in Study 2. The study was a parallel evaluation (N=50) of intranasal insulin (60 IU) vs placebo. Participants were tested after 16 hours of smoking abstinence. As in Study 1, intranasal insulin had no effect on learning (trial 1-5), trial B (distractor trial), or any of the 4 recall trials as listed in the figure.

Table 1

Demographic characteristics of study participants

	Intranasal Insulin (n=26)	Placebo (n=24)
<i>Characteristics</i>		
Age, mean (s.d.)	36.5 (11.82)	38.30 (13.33)
Gender (Male)	80.77%	66.67%
Race (White)	84.62%	100.00%
Hispanic ethnicity	23.08%	37.50%
Education		
12th grade or less	11.54%	0.00%
High school graduate	23.08%	29.17%
Some college/AA degree	53.85%	50.00%
College Graduate	7.69%	20.83%
Graduate school degree	3.85%	0.00%
Unemployed	26.92%	37.50%
Fagerstrom Test for Nicotine Dependence, mean (s.d)	4.38 (2.62)	5.42 (1.89)
Smoking quantity (cigarettes per day), mean (s.d.)	16.15 (7.56)	18.50 (5.76)
Age started smoking, mean (s.d.)	16.60 (4.09)	16.69 (3.19)
Body mass index, mean (s.d.)	25.52 (3.21)	25.33 (3.24)
Alcohol use (last 30 days)	63.64%	60.47%
Marijuana use (last 30 days)	10.53%	13.33%
Illicit Drug Use (ever)		
Cocaine	45.83%	36.36%
Heroin	4.17%	8.70%
Tranquilizers	4.17%	4.35%
Pain Relievers	29.17%	8.70%
Amphetamines	16.67%	30.43%
Hallucinogens	54.17%	43.48%

Table 2

Result of Learning Analysis (Study 1)

PARAMETER	COEFFICIENT	STANDARD ERROR	z	p	95% CONFIDENCE INTERVAL	
Treatment	-0.022	0.015	-1.520	0.129	-0.051	0.006
Trial	-0.048	0.004	-11.840	0.001	-0.056	-0.040
Treatment × Trial	0.008	0.006	1.430	0.151	-0.003	0.019
Order of Treatment	-0.007	0.027	-0.260	0.796	-0.059	0.045

Table 3

Results of Memory Analyses (Study 1)

	PARAMETER	COEFFICIENT	STANDARD ERROR	z	p	95% CONFIDENCE INTERVAL	
TRIAL B	Treatment	0.175	0.480	0.360	0.715	-0.766	1.116
	Order of Treatment	0.992	0.659	1.500	0.133	-0.301	2.284
SHORT DELAY FREE RECALL	Treatment	-0.034	0.048	-0.710	0.478	-0.128	0.060
	Order of Treatment	-0.127	0.116	-1.090	0.277	-0.355	0.101
SHORT DELAY CUED RECALL	Treatment	-0.057	0.066	-0.870	0.384	-0.187	0.072
	Order of Treatment	-0.149	0.113	-1.320	0.188	-0.372	0.073
LONG DELAY FREE RECALL	Treatment	-0.048	0.077	-0.620	0.537	-0.200	0.104
	Order of Treatment	-0.250	0.185	-1.350	0.177	-0.613	0.113
LONG DELAY CUED RECALL	Treatment	-0.111	0.121	-0.920	0.360	-0.349	0.127
	Order of Treatment	-0.207	0.208	-0.990	0.320	-0.614	0.201

Table 4

Result of Learning Analysis (Study 2)

PARAMETER	COEFFICIENT	STANDARD ERROR	z	p	95% CONFIDENCE INTERVAL	
Treatment	0.016	0.020	0.810	0.420	-0.023	0.055
Trial	0.040	0.003	12.720	0.001	0.034	0.046
Treatment × Trial	-0.006	0.005	-1.270	0.206	-0.015	0.003

Table 5

Results of Memory Analyses (Study 2)

PARAMETER	COEFFICIENT	STANDARD ERROR	z	p	95% CONFIDENCE INTERVAL	
TRIAL B	-0.041	0.087	-0.470	0.635	-0.211	0.129
SHORT DELAY FREE RECALL	0.048	0.060	0.800	0.423	-0.070	0.166
SHORT DELAY CUED RECALL	0.015	0.107	0.140	0.891	-0.195	0.224
LONG DELAY FREE RECALL	0.028	0.082	0.340	0.735	-0.134	0.189
LONG DELAY CUED RECALL	0.012	0.133	0.090	0.929	-0.248	0.272