

**Society of Behavioral Medicine  
Distinguished Scientist Master Lecture**

**The Value of Animal Studies to  
Understand and Treat Tobacco Use**

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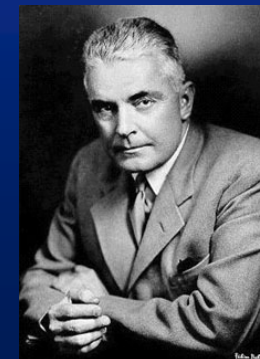
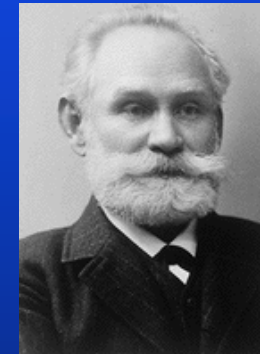
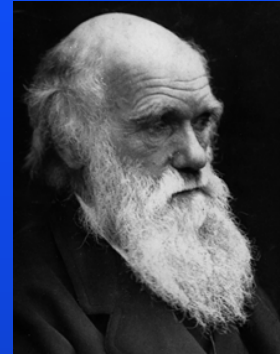


# Overview

- Psychology and the value of animal research
- Behavioral medicine and the value of animal research
- Parallel human and animal research to address an important topic in behavioral medicine: tobacco use
- Translational research in behavioral medicine

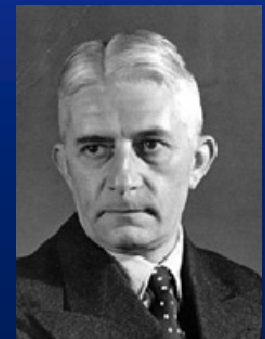
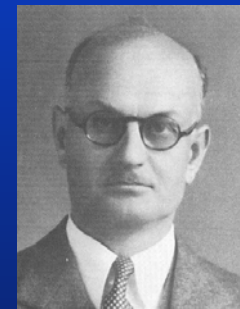
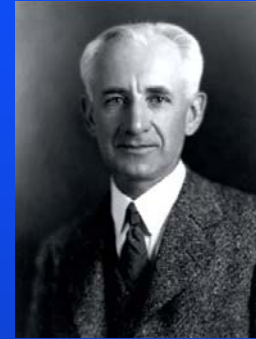
# Psychology and Animal Research

- Darwin (1809 -1882)
  - Postulated continuity of species
  - Argued that survival is determined by organism's ability to adapt to its environment
- Pavlov (1849 - 1936)
  - Studied GI physiology and serendipitously “discovered” classical conditioning
- Thorndike (1874 -1949)
  - Studied learning in cats and discovered “Law of effect “
- Watson (1878 -1958)
  - Extended reinforcement principles
  - Studied neurons and learning in rats



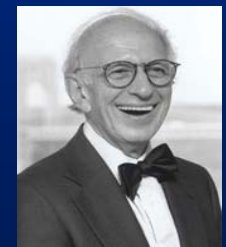
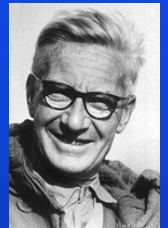
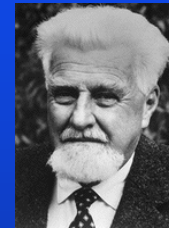
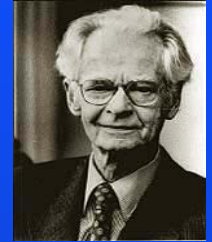
# Psychology and Animal Research

- **Yerkes** (1876 - 1956)
  - Studied learning and motivation in animals
- **Lashley** (1890 - 1958)
  - Studied effects of brain damage on learning and memory in animals
- **Tolman** (1886 - 1959)
  - Studied “cognitive maps” in rats
- **Köhler** (1887 - 1967)
  - Studied cognitive processing in animals



# Psychology and Animal Research

- Skinner (1904 - 1990)
  - Extended Thorndike's and Watson's work to develop instrumental learning and operant conditioning
- Lorenz & Tinbergen (1903 -1989/ 1907 -1988)
  - Studied behavior of animals in their natural environments (ethology)
- Hebb (1904 -1985)
  - Studied neurons during psychological functioning (neuroplasticity)
- Rosenzweig (1922 - )
  - Studied psychological and biological effects of enriched environments on rats
- Kandel (1929 - )
  - Studied neurobiological basis of learning and memory in *Aplysia*



# Behavioral medicine & animal research

- **Miller** (1909 - 2002)
  - Studied cognitive control of autonomic nervous system (mind/body) and developed biofeedback
  - Studied approach/avoidance situations to understand psychological conflict
  - Studied biological bases of stress and mental health
- **Selye** (1907 - 1982)
  - Studied stress, perceived control, and pathophysiological consequences
- **Mason** (1925 - )
  - Studied stress, perceived stress, perceived control
- **Brady** (1922 - )
  - Studied perceived control, stress, and health consequences

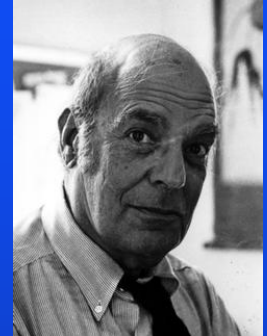


# Behavioral medicine & animal research

- **Stress:** *e.g.*, Brady, Cannon, Mason, Selye, Singer
- **CVD:** *e.g.*, Cox, Harvey, Henry, Herd, Hubbard, Lawler, Manuck, Schneiderman
- **Diabetes:** *e.g.*, Cannon, Surwit, Woods
- **PNI and Cancers:** *e.g.*, Ader, Maier
- **Pain:** *e.g.*, Melzack, Wall, Yaksh
- **Obesity:** *e.g.*, Hirsch, Greenwood, Stellar, Surwit
- **Tobacco/Nicotine:** *e.g.*, Caggiula, Corrigall, Damaj, Domino, Donny, Goldberg, Grunberg, Jarvik, Levin, Malin, Picciotto, Stolerman

# Combining Social Psychology, Psychobiology, and Pharmacology

- Schachter (1922 – 1997)
  - Theory-driven research regarding social comparison: affiliation, anxiety, hunger
  - Problem-oriented research: obesity, cigarette smoking
- Grunberg research (beginning late 1970s)
  - **Why do people who quit smoking gain weight?**
  - Include human and animal laboratory experiments to address clinical behavioral medicine question (inspired by work of Neal E. Miller and by doctoral dissertation of Jerome E. Singer)
  - Research designed to examine clinically-relevant question and to reveal basic biobehavioral mechanisms (including potential role of nicotine and hypothesized relationships among drug actions and appetitive behaviors)



# Cigarette Smoking and Body Weight

- Review of literature:
  - Inverse relationship between tobacco use and body weight
  - Three suggested mechanisms:
    - Oral gratification
    - Changes in food consumption (general or specific)
    - Changes in metabolism
- Human laboratory experiment
  - Smokers smoking, smokers not smoking, and nonsmokers not smoking
  - General and specific food consumption
- Animal laboratory experiment (rats)
  - Administering nicotine subcutaneously (SC)
  - General and specific food consumption
  - Body weight

# Value of Animal Models to Examine Human Smoking

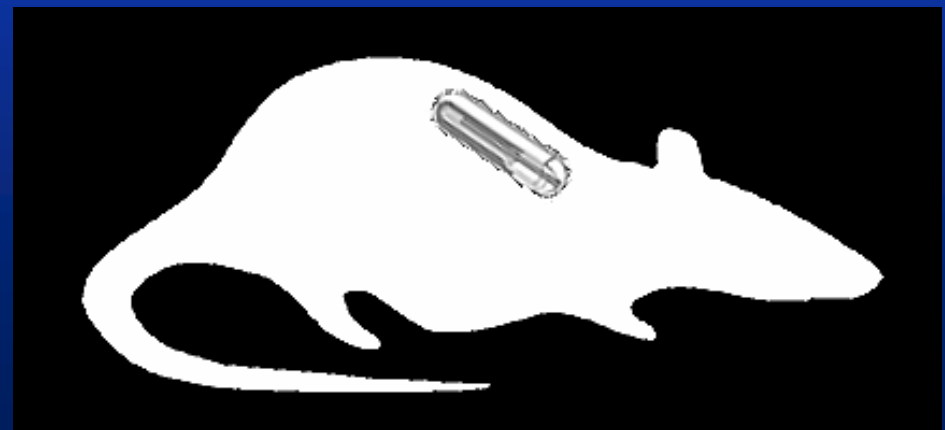
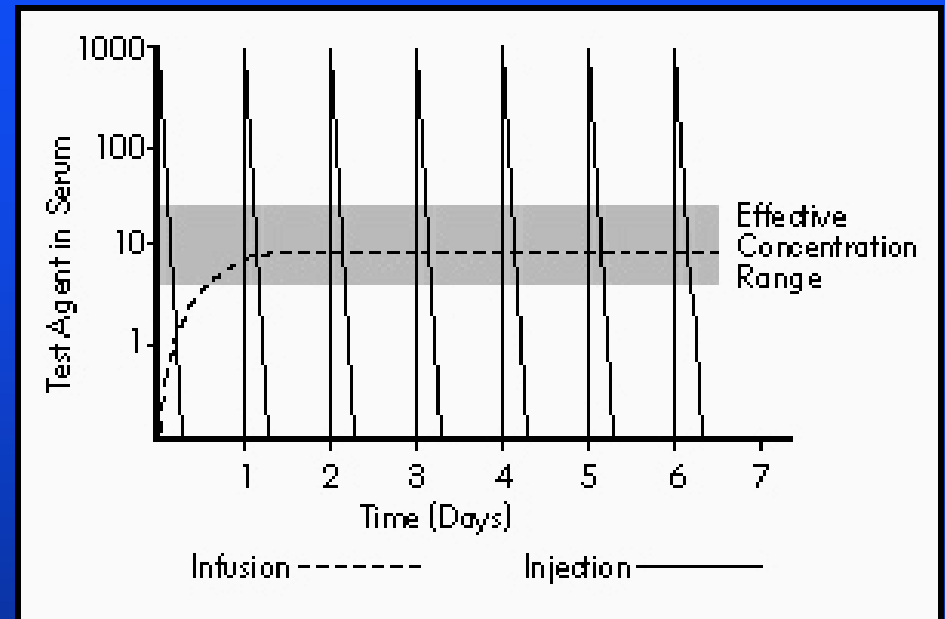
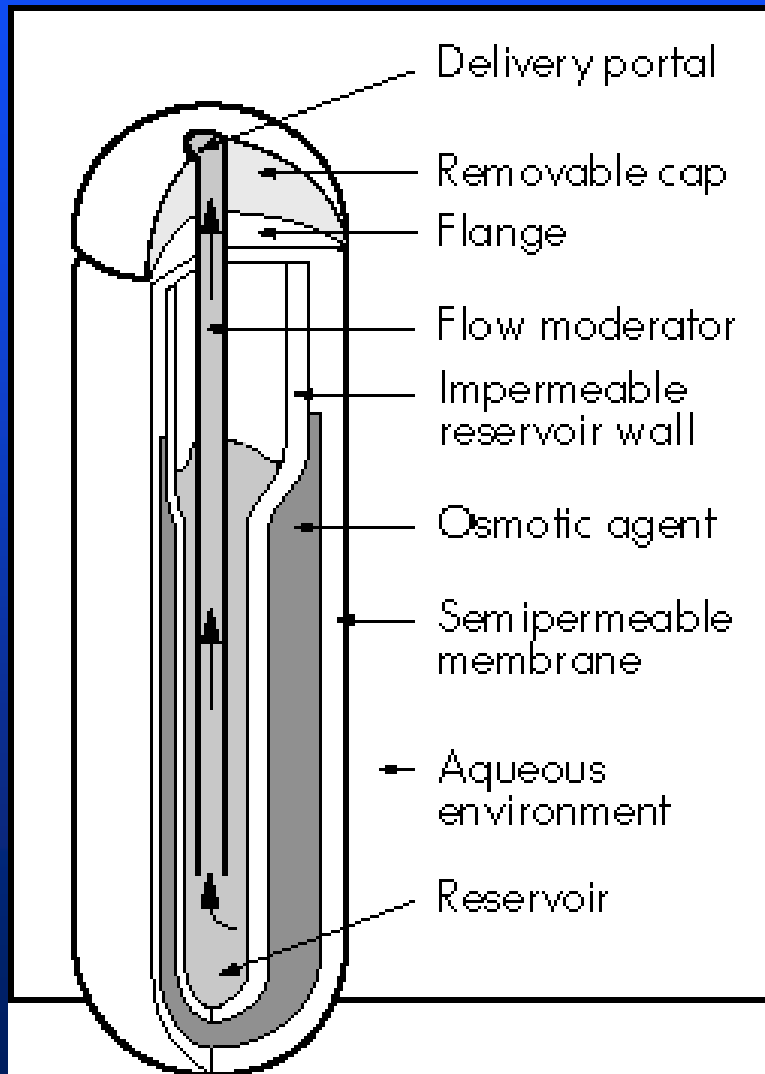
- **Conceptual**
  - Separation of biological and psychosocial factors
  - Findings parallel and predict findings from human studies
- **Methodologic**
  - Experimental control
  - Establish causality
- **Ethical**



# How to administer nicotine

- Hoped to use self-administration paradigm but...
- Alzet osmotic minipump (thanks to H. Hansen)
- Minipump implanted SC (0 – 12 mg/kg/day)
- Evaluated during nicotine administration (weeks) and during cessation (weeks)
- Selected to:
  - rule out “oral gratification” explanation
  - evaluate nicotine hypothesis
  - to “model” tobacco use while avoiding technical challenges of self-administration paradigm

# Osmotic minipump



# Initial findings regarding tobacco/nicotine and body weight

- Nicotine attenuated body weight gains
- Cessation of nicotine resulted in increased weight gains
- Tobacco use (by humans) and nicotine (in rats) decreased sweet food consumption
- Abstinence from tobacco and cessation of nicotine increased sweet food consumption

# Subsequent Research in our Lab: Tobacco/Nicotine and Body Weight

- Scientific Questions:
  - Does nicotine reduce body weight? What are the biological and behavioral effects of nicotine that contribute to changes in body weight? Are there sex and age differences in these effects?
- Clinical Questions:
  - How can we help people who quit smoking avoid weight gains?
  - Are there individual differences (based on sex, age, or genetics) in the relationship between tobacco use and body weight?
- Research conducted from 1980 - present:
  - Cigarette smoking, body weight, and food consumption (humans)
  - Nicotine, body weight, food consumption, physical activity, and metabolism (rats)
  - Examination of individual differences using various nicotine administration paradigms in rats

# Nicotine and Body Weight: Design (one experimental example)

- **Subjects**

- 80 SD rats
  - 40 males and 40 females
  - half adolescent (30 days old) and half adult (65 days old)



- **Design**

- 2 (male or female) X 2 (adolescent or adult) X 2 (saline or 12 mg/kg/day nicotine)

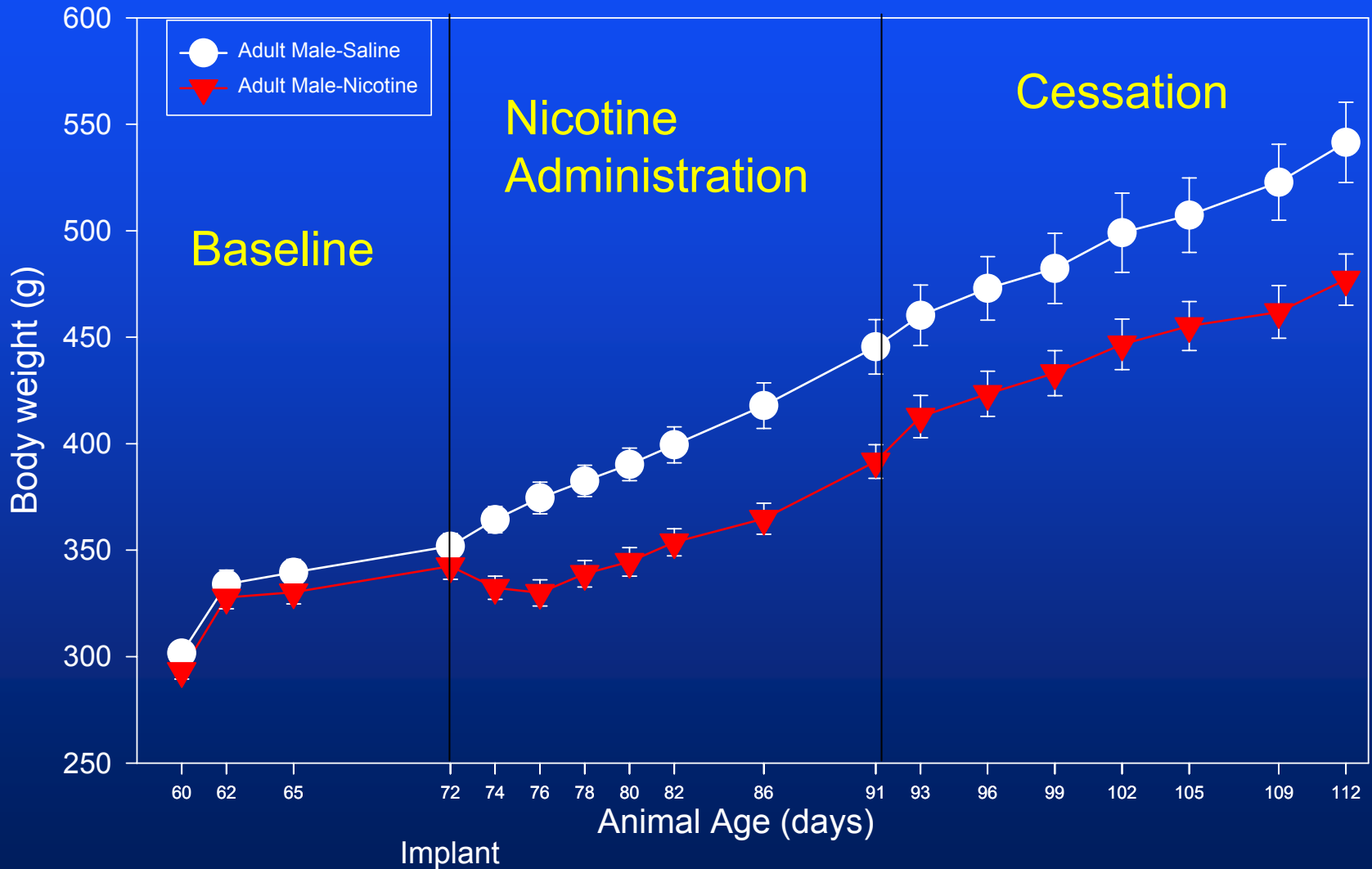
- **Nicotine administration**

- via osmotic mini-pump for 3 wks

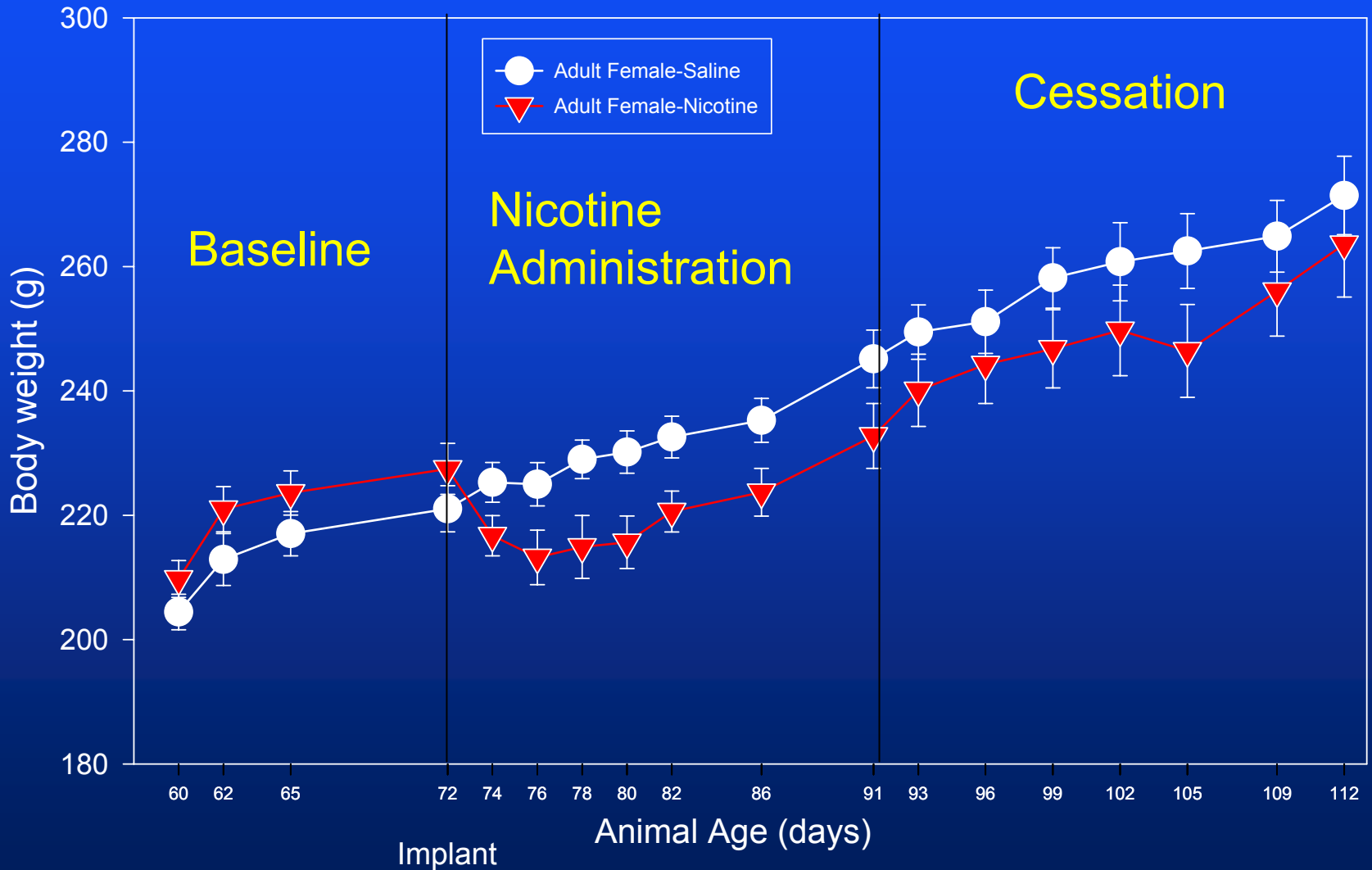
- **Dependent Measures**

- Body weight & food consumption (bland chow)

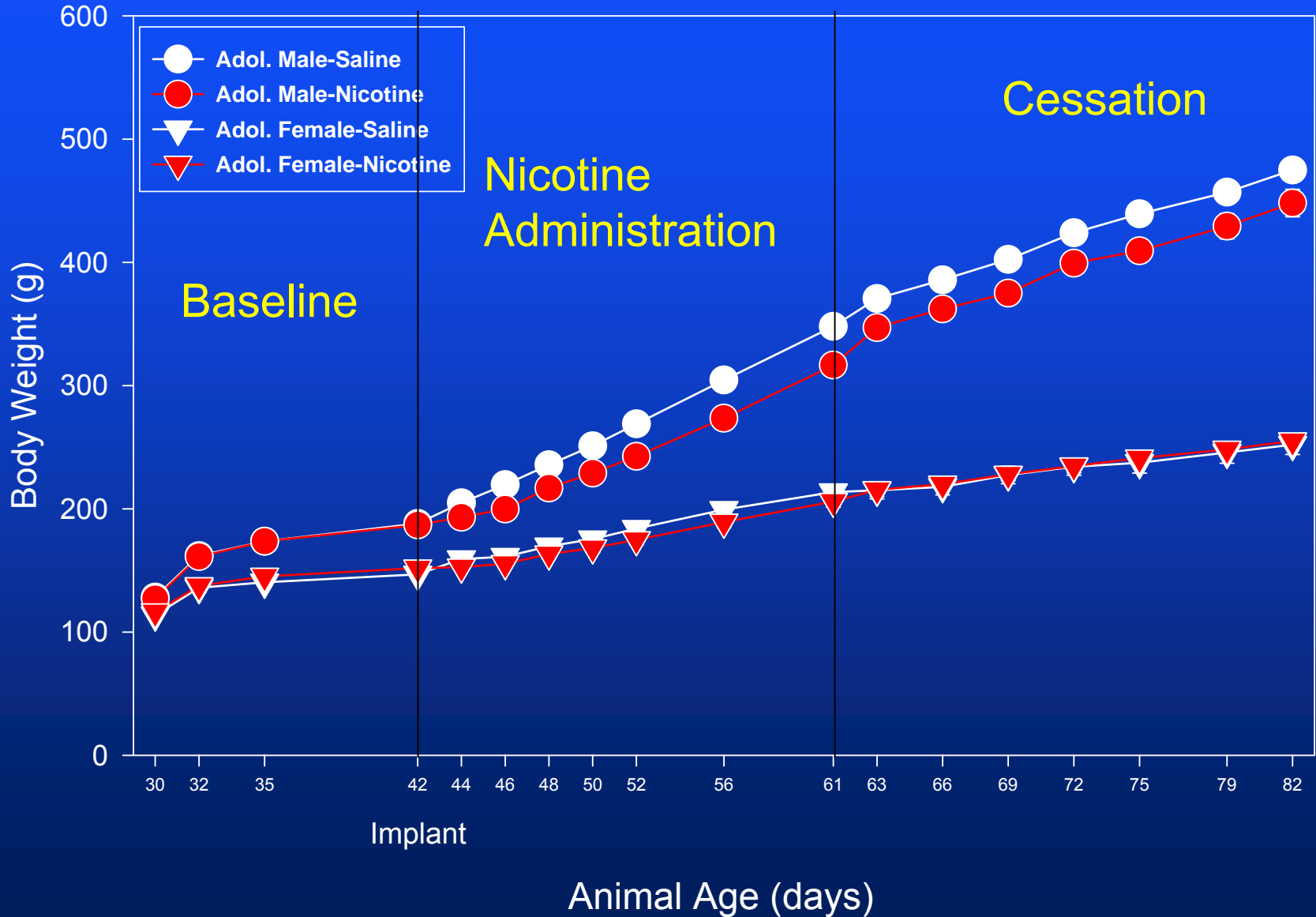
# Results: Adult Males Body Weight



# Results: Adult Females Body Weight



# Results: Adolescent Body Weights



# Nicotine and Body Weight: Results

## Adults

- Nicotine **suppressed** body weight gain and feeding in **adult males and females**, consistent with previous findings

## Adolescents

- Nicotine **suppressed** feeding and body weight gain of **adolescent males**
- Nicotine **did not alter** feeding and body weight of **adolescent females**

# Nicotine and Body Weight: Findings from Additional Experiments

- Nicotine suppresses body weight and feeding (especially high caloric, sweet foods) in adult males and females
  - Effect in females > Effect in males
- Cessation of nicotine increases body weight gains and feeding in adult males and females
  - Effect in females > Effect in males
- Nicotine reduces body weight and feeding in adolescent males, but not adolescent females
  - Effect in males > Effect in females
- Cessation of nicotine somewhat increases body weight gains and feeding in adolescent males but not females
  - Effect in males > Effect in females

# Nicotine and Body Weight: Clinical Implications

- To avoid weight gain during smoking cessation strategies should include decreasing consumption of high caloric sweet foods
  - For men and women
  - High caloric sweet foods can be replaced with low-caloric sweet foods
- Adolescent girls should be aware that smoking cessation has little impact on weight gain

# Additional research topics regarding tobacco use and treatment

- Body weight
- Stress
- Attention
- Pain
- Sex differences
- Age differences
- Genetic differences
- Environmental effects
- Withdrawal

# Nicotine and Stress

- Smokers smoke more under stress (Schachter et al., 1977)
- Urinary pH as mediator of increased nicotine elimination (hypothesized pharmacokinetic mechanism)
- “pH monkey” study
  - pH manipulated by IG infusion of acidifier and alkalinizer
  - Behavioral effects of nicotine under pH conditions evaluated on multiple FI-FR stimulus-shock termination schedule evaluated
  - Results indicated pharmacodynamic effect
- Theme recurred in the lab with rabbit and rat studies (e.g., D. Morse, S. Winders-Barrett) that revealed PK effects as well

# Nicotine and Anxiety

- Scientific Question:
  - Does nicotine alter stress and anxiety?
- Clinical Question:
  - How to help people cope with stress? How to help people who use tobacco to cope with stress find alternative stress-coping techniques?

# Nicotine and Anxiety: Design (one example)

- **Subjects**

- 160 SD rats
  - 80 males and 80 females
  - half adolescent (30 days old) and half adult (65 days old)



- **Design**

- 2 (adolescent or adult) X 4 (saline, 0.10, 0.50 or 1.0 mg/kg nicotine) X 2 (male or female); n = 10 per cell

- **Nicotine administration**

- via daily SC injection for 5 days

- **Dependent Measures**

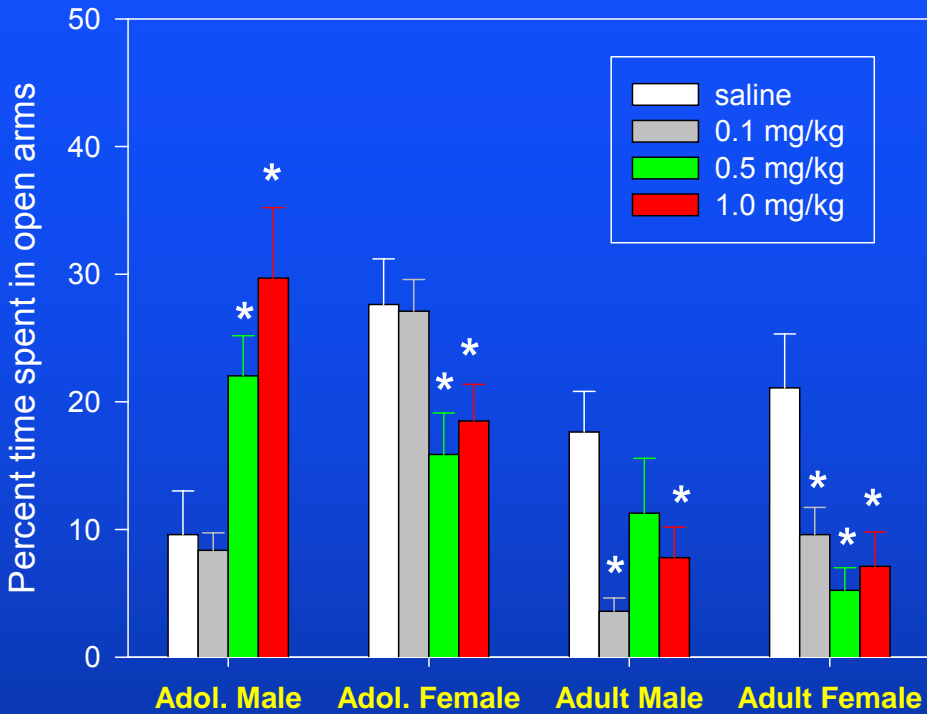
- Elevated Plus maze for 5 min, 10 min post injection

# Elevated Plus-Maze Activity

- Behavioral measure of anxiety in rodents
- Based on principle that animals find open areas aversive
- Widely-used for drug screening



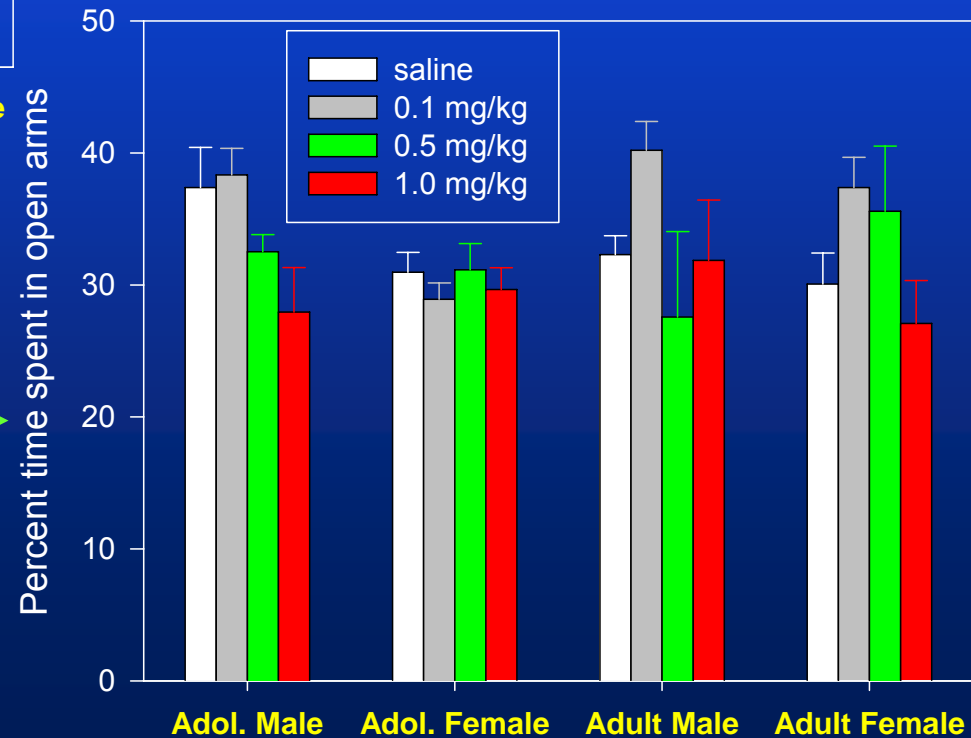
## Percent Time Open



← Measure of anxiety

Measure of activity →

## Percent Entries Closed



# Nicotine and Anxiety: Results

## Adults

- Nicotine **increased** an index of anxiety in **adult males and females** (anxiogenic in adults)

## Adolescents

- Nicotine **decreased** an index of anxiety in **adolescent males** (anxiolytic)
- Nicotine **increased** an index of anxiety in **adolescent females** (anxiogenic)

# Nicotine and Anxiety: Clinical Implications

- For adolescent males, tobacco prevention and treatment strategies should focus on non-tobacco coping strategies for anxiety reduction
- For adolescent females and adults, perceptions that nicotine may reduce anxiety should be challenged

# Nicotine and Attention

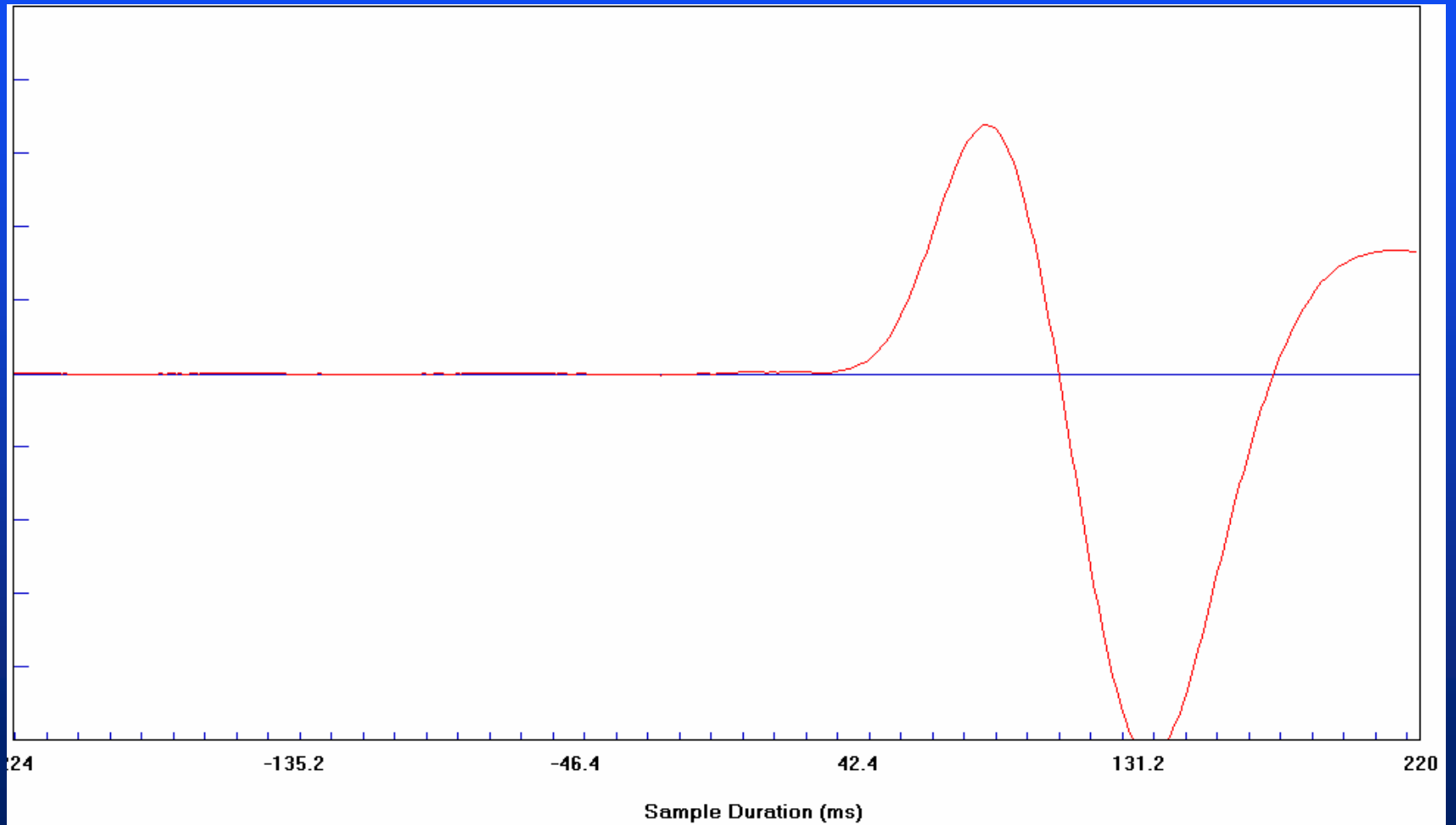
- Scientific Question
  - Does nicotine improve sensorimotor gating and information processing?
- Clinical Question
  - Does nicotine improve attention?

# Pre-pulse inhibition of acoustic startle response

- Procedure (acoustic startle reflex [ASR])
  - Animal placed in sound-attenuated chamber
  - Auditory stimulus (i.e., 120 dB) presented to animal
  - Animal startles to noise



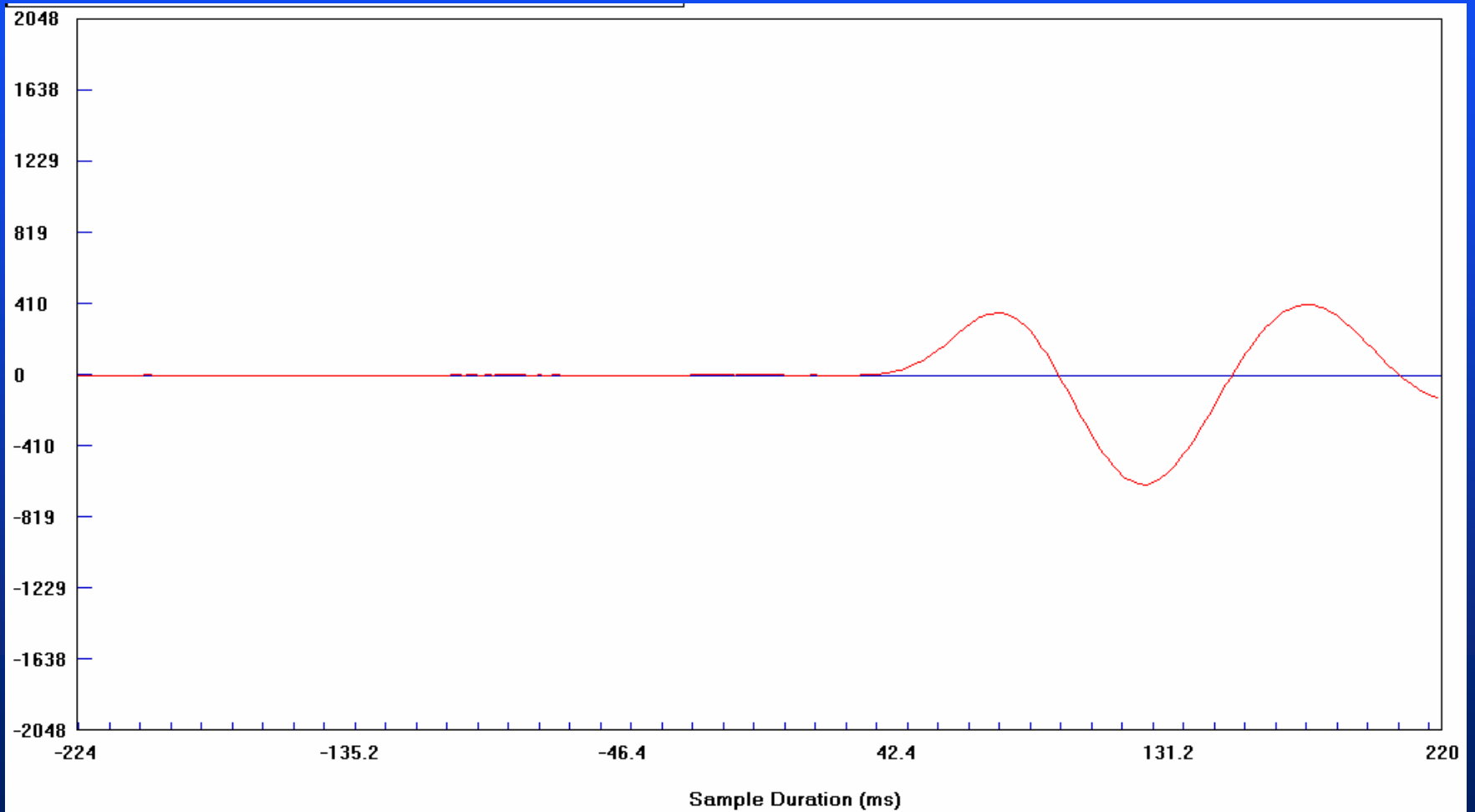
# Acoustic Startle Reflex



# Pre-pulse inhibition of acoustic startle response

- Procedure (pre-pulse inhibition)
  - On some trials stimulus is preceded by a non-startling stimulus (*i.e.*, 82 db)
  - Animal's response to louder stimulus is again recorded
  - Response to stimulus should be reduced when preceded by non-startling stimulus

# Pre-pulse Inhibition



# Attention: pre-pulse inhibition of acoustic startle

- Interpretation
  - Impairments in pre-pulse inhibition reflect impairments in information processing, sensorimotor gating, and attention
- Purpose
  - Used to index effects of drugs on attention
  - Used to index attentional problems that characterize various disorders (e.g., Schizophrenia, ADHD)

# Nicotine and Attention: Design (one example)

- **Subjects**

- SD male rats



- **Design**

- 3 (pre/during/post drug) X 3 (saline, 6, 12 mg/kg nicotine)

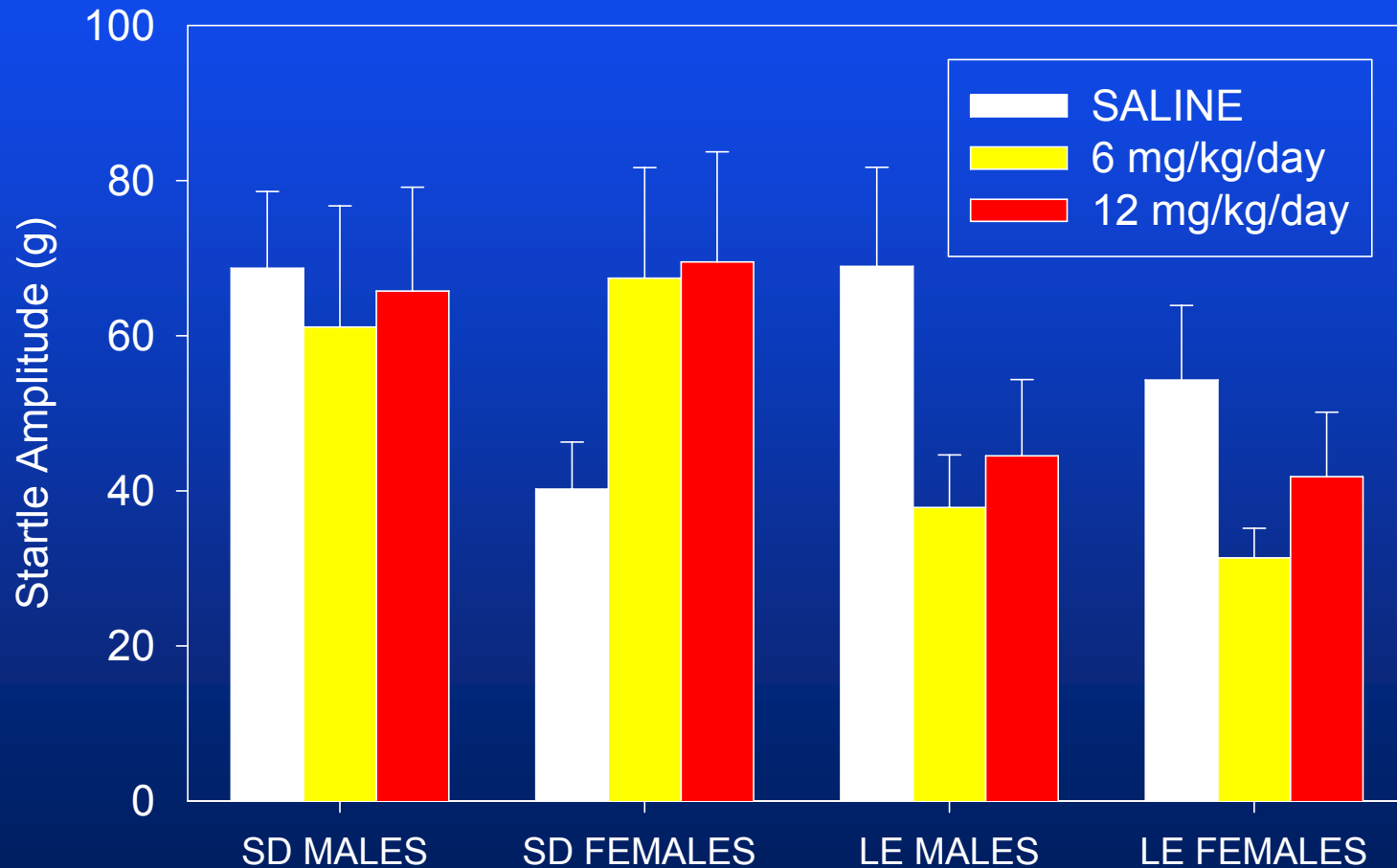
- **Nicotine administration**

- via osmotic mini-pump for 10 days

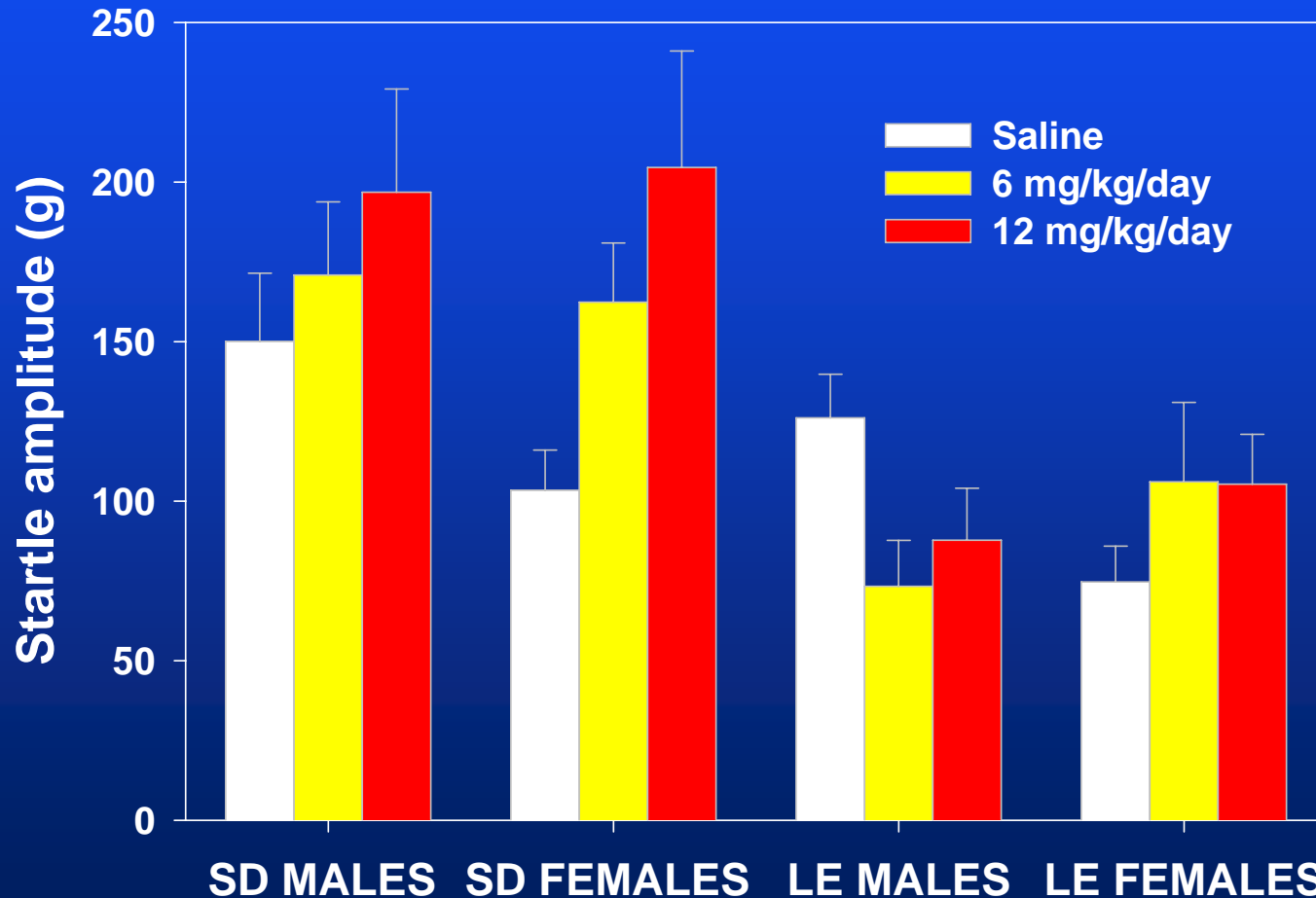
- **Dependent Measures**

- Acoustic startle response
- PPI of ASR

# Startle Amplitude after Two Days of Nicotine Administration



# Startle Amplitude after Six Days of Nicotine Administration



# Nicotine and Attention: Results

- Nicotine enhanced ASR and PPI in SD rats
  - These effects appeared earlier and across more stimuli for females compared with males
- These effects decreased during cessation
- Effects are marked and significant in adult rats
- Minimal effects in adolescent rats
- Genetic differences exist

# Nicotine and Attention: Clinical Implications

- Nicotine helps to increase selective attention, sustained attention, and vigilance, and also decreases effects of distracters
- It is important to teach patients non-tobacco coping strategies for increasing attention
- Nicotine replacement therapy (NRT) may be useful to improve attention in adults

# Nicotine and Pain

- Scientific Question:
  - Does nicotine affect nociception?
- Clinical Question:
  - Do some individuals smoke to relieve pain?

# Nicotine and Pain: Design

- **Subjects**

- 300 Sprague Dawley rats
  - 150 males and 150 females
  - half adolescent (30 days old) and half adult (65 days old)



- **Design**

- 2 (adolescent or adult) X 5 (saline, 0.01, 0.10, 0.50 or 1.0 mg/kg nicotine) X 2 (male or female)

- **Nicotine administration**

- via daily SC injection for 10 days

- **Dependent Measures**

- Hot Plate measured on days 2 or 3 and 8 or 9
- Tail Flick measured on days 2 or 3 and 8 or 9

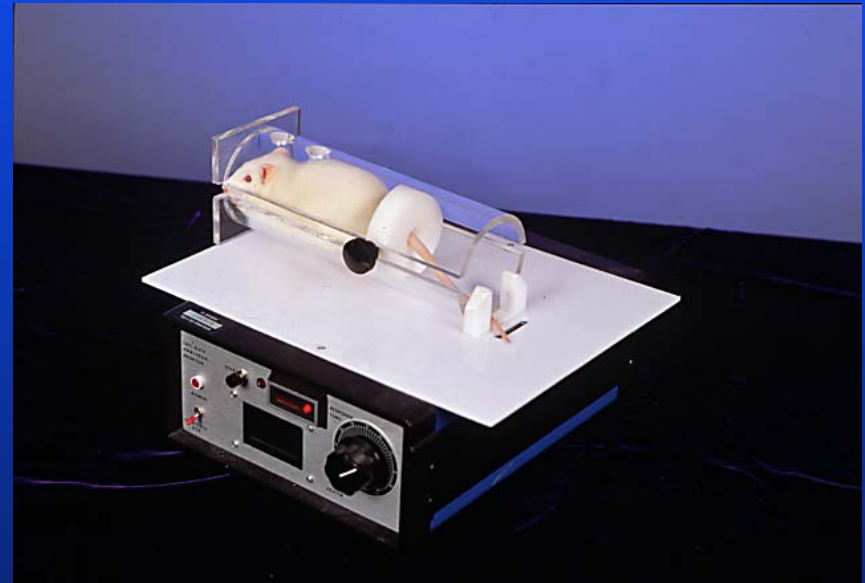
# Hotplate

- Hotplate is a supraspinally-mediated measure of pain
- Measured on days 2 or 3 and 8 or 9
- Animals tested at 8 and 12 minutes post-injection

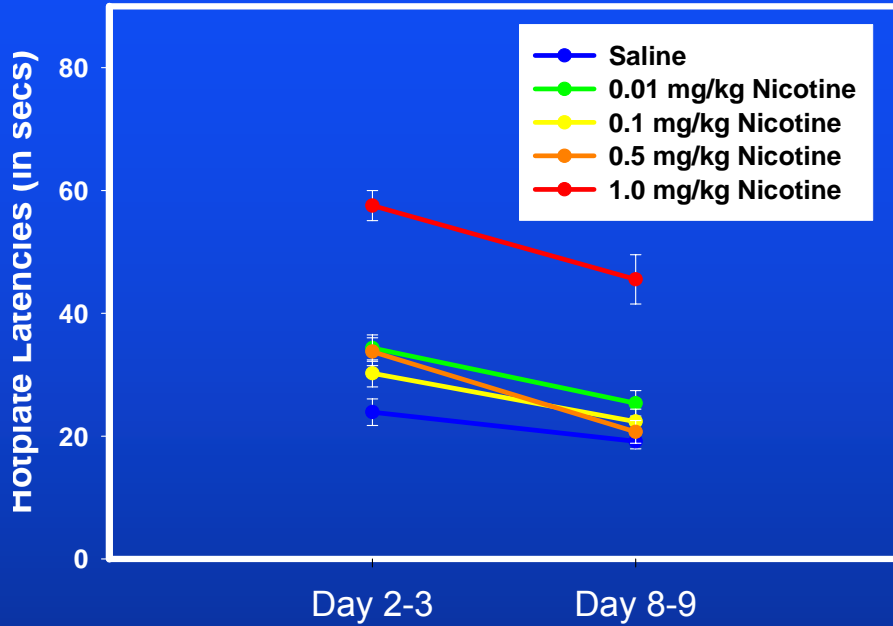


# Tail Flick

- Tail flick is a spinally-mediated measure of pain
- Measured on days 2 or 3 and 8 or 9
- Animals tested at 8 and 12 minutes post-injection

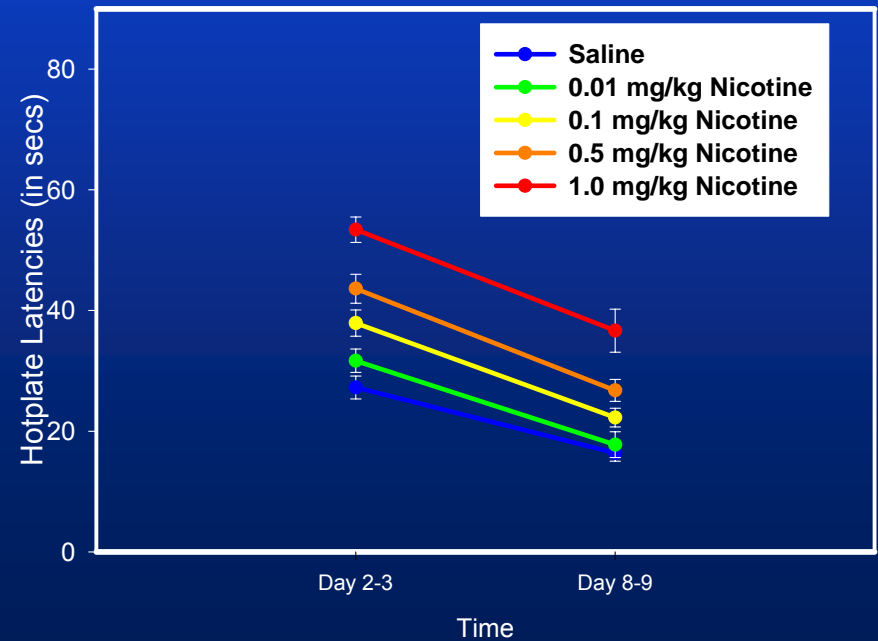


# Hotplate Results: Adults

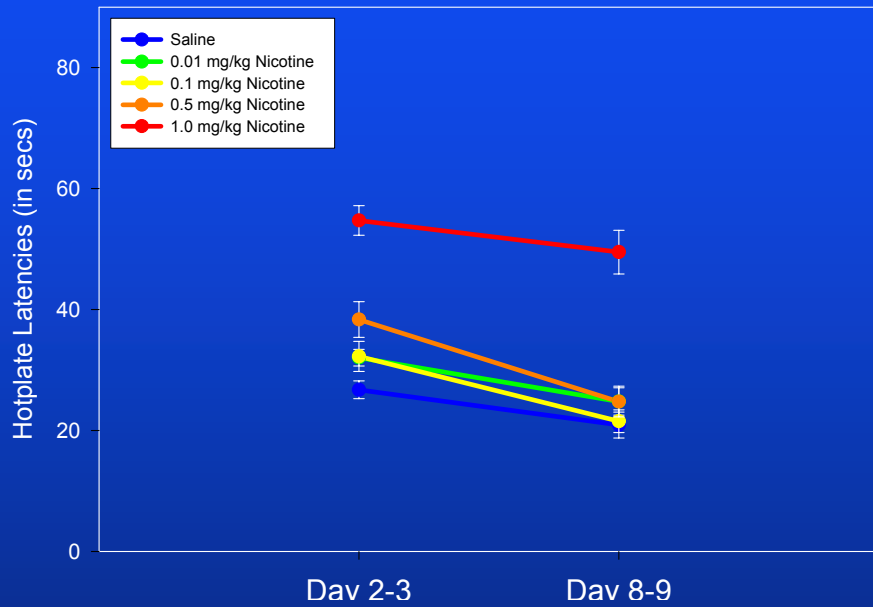


← **MALES**

**FEMALES** →



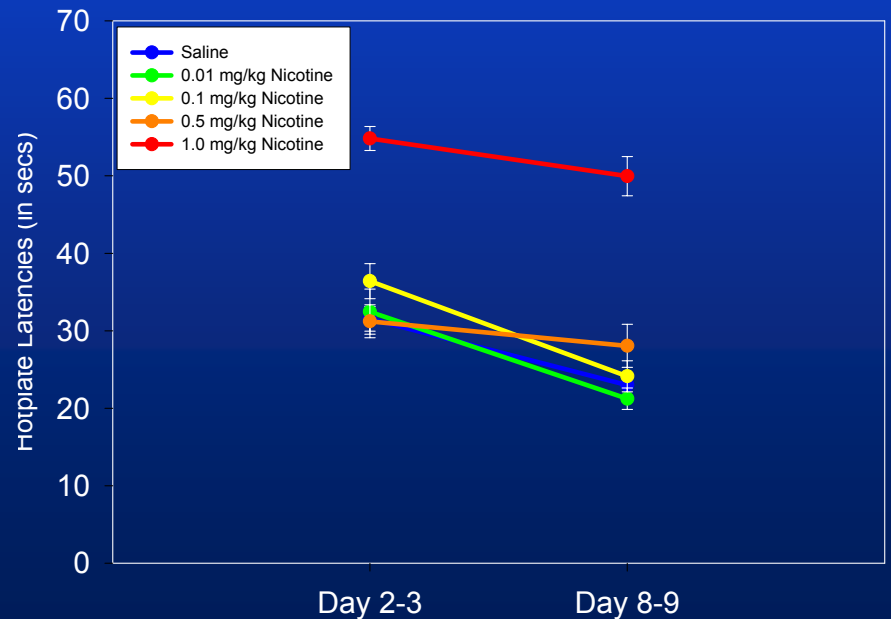
# Hotplate Results: Adolescents



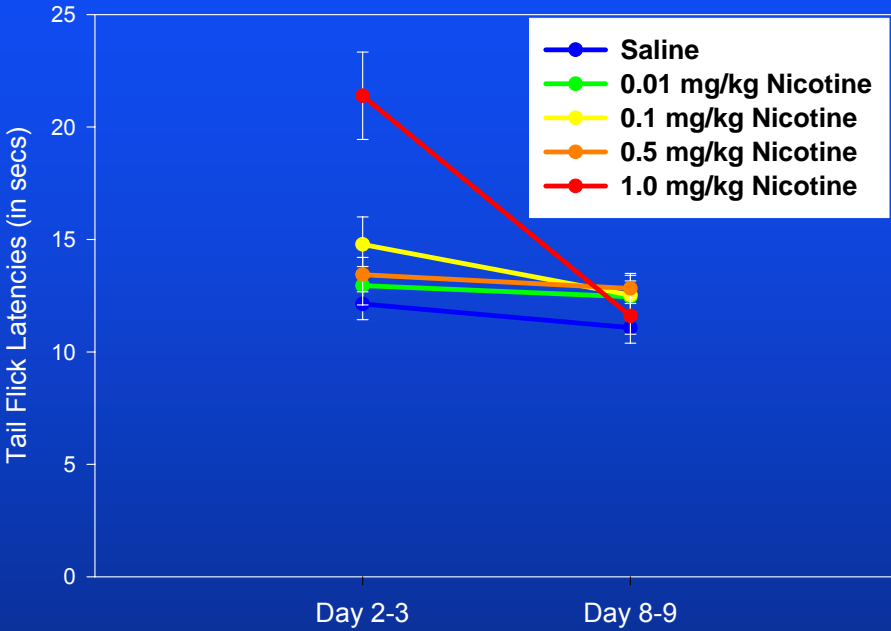
**FEMALES**



**MALES**

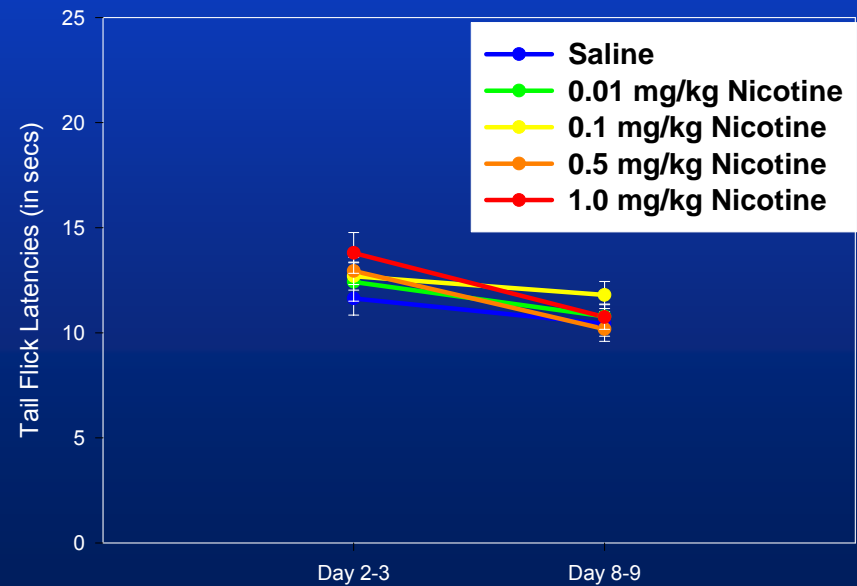


# Tail Flick Results: Adults

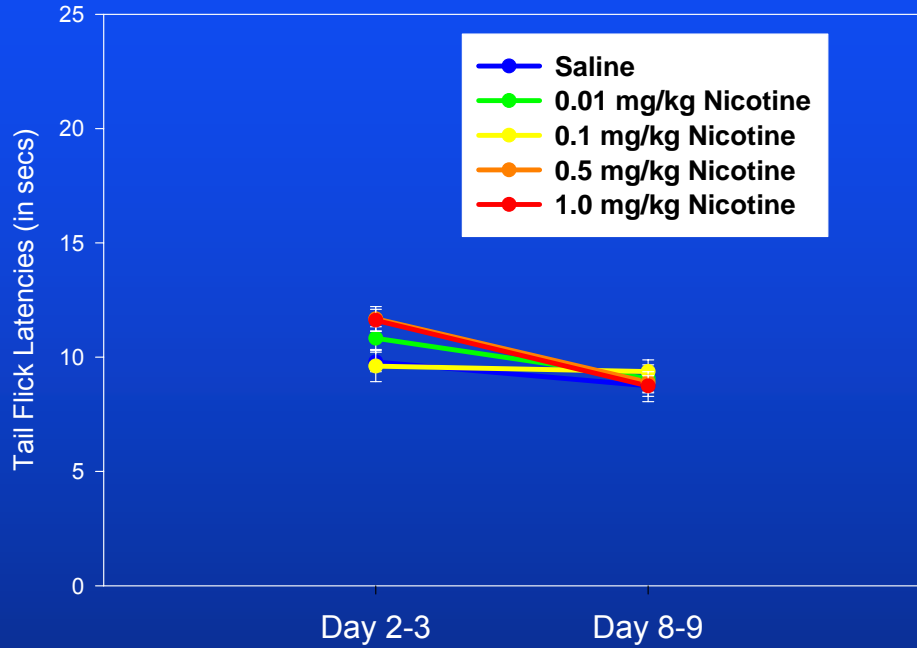


**MALES**

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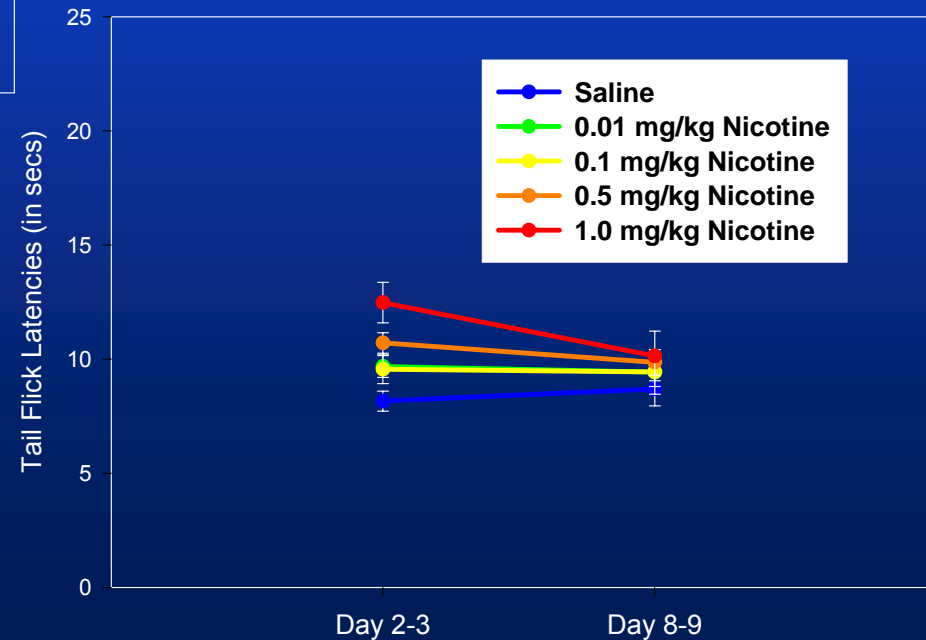


# Tail Flick Results: Adolescents

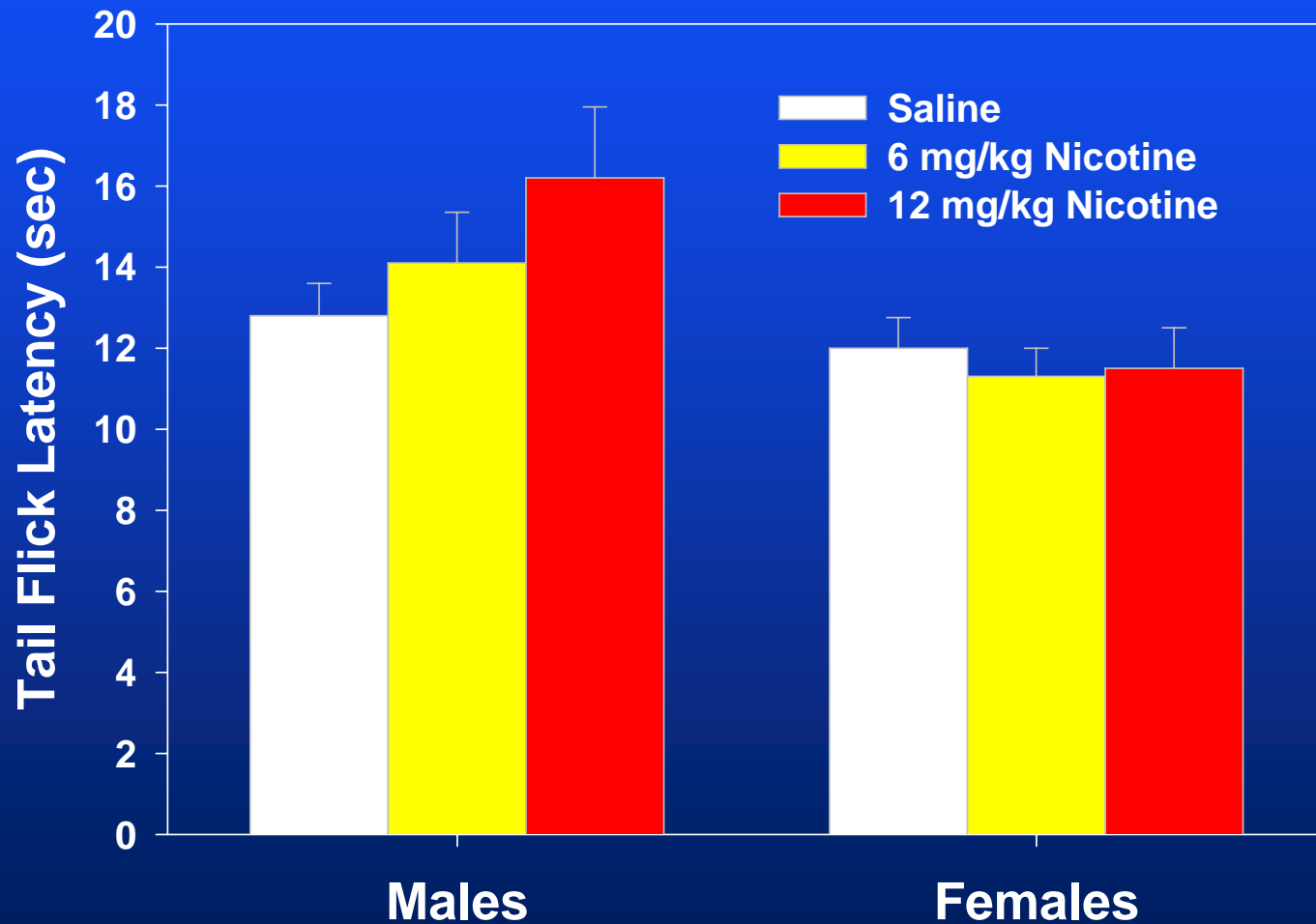


← MALES

FEMALES →



# Nicotine and Anti-nociception: Sprague-Dawley Males vs. Females



# Nicotine and Pain: Results

- Nicotine significantly decreased nociception for all groups
- **Hotplate**
  - Adults were more sensitive than adolescents
  - Nicotine **increased** analgesia in adult males and females
- **Tail-flick**
  - Adult males were most sensitive compared with adult females and adolescents
  - Nicotine **increased** analgesia in male rats and had **no effect** in female rats

# Nicotine and Pain: Clinical Implications

- **Antinociceptive effects** may be especially important for **adult males**
- Pain should be assessed prior to smoking cessation and pain coping techniques should be taught as needed

# Nicotine and Environment

- Scientific Question:
  - Do environmental conditions alter nicotine's effects? Does physical and social enrichment alter nicotine's effects?
- Clinical Question:
  - Do differences in the environment lead to different levels of sensitivity of nicotine? Could environmental differences (e.g., socioeconomic status, culture) in tobacco use be explained by altered responses to nicotine?

# Nicotine and Environment: Design (one example)

- **Subjects**

- 72 Sprague Dawley **male adolescent** rats



- **Design**

- 4 (saline or 0.10, 0.50, 1.0 mg/kg nicotine) X 3 (isolated, social, or enriched); n = 6 per tx group
- Later replicated with **female rats**

- **Nicotine administration**

- via daily SC injection for 16 days

- **Dependent Measures**

- Locomotion measured on days 2, 6, and 9

# Environments

Isolated Environment



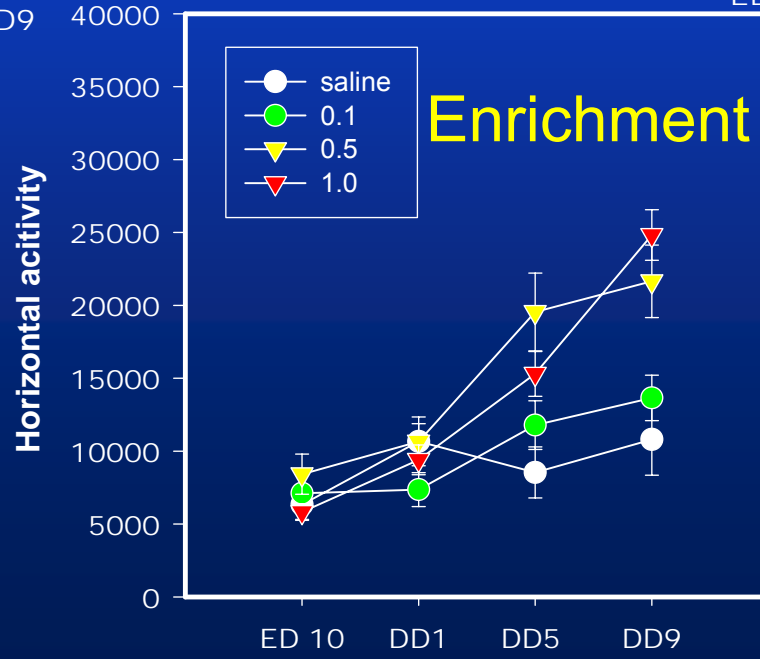
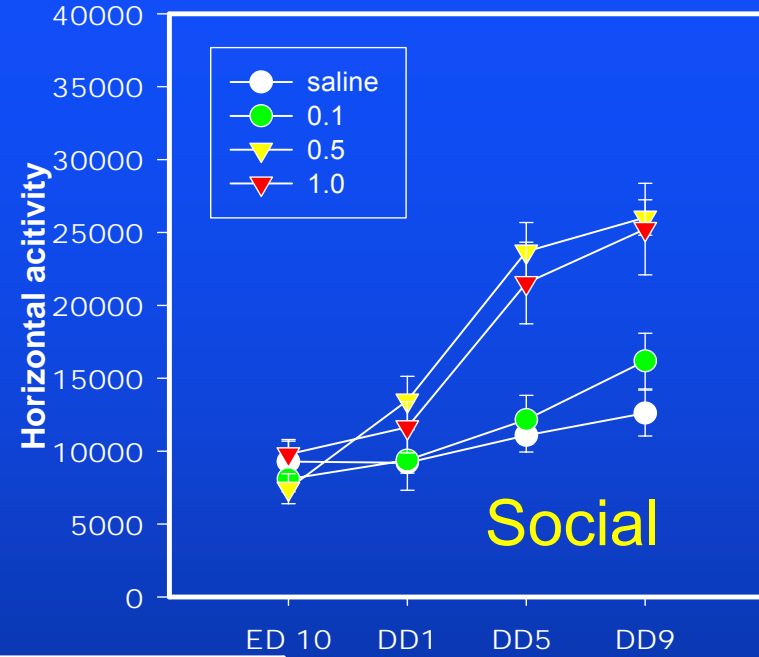
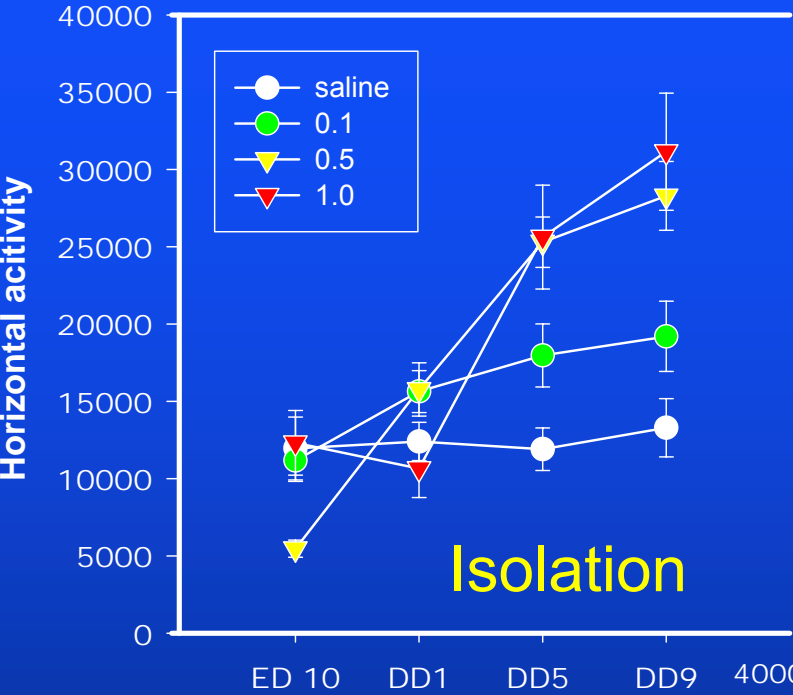
Social Environment



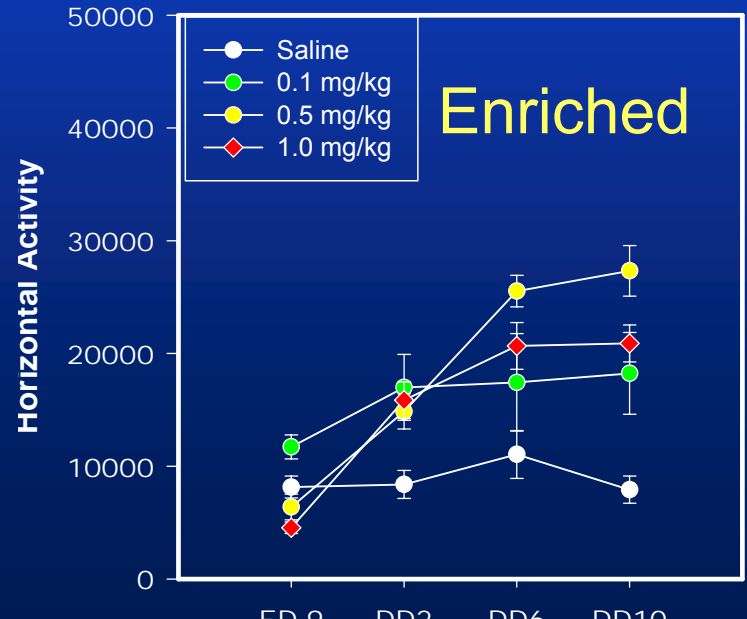
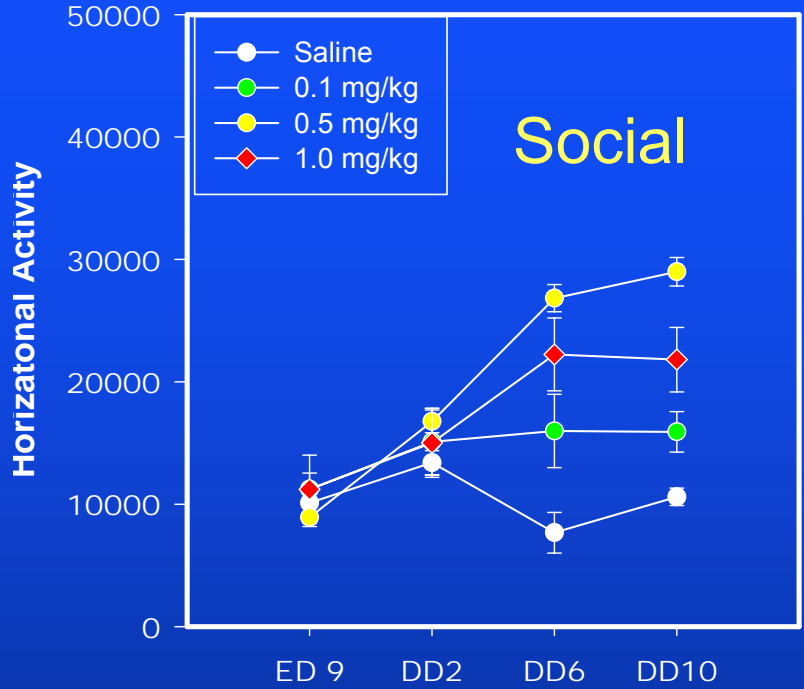
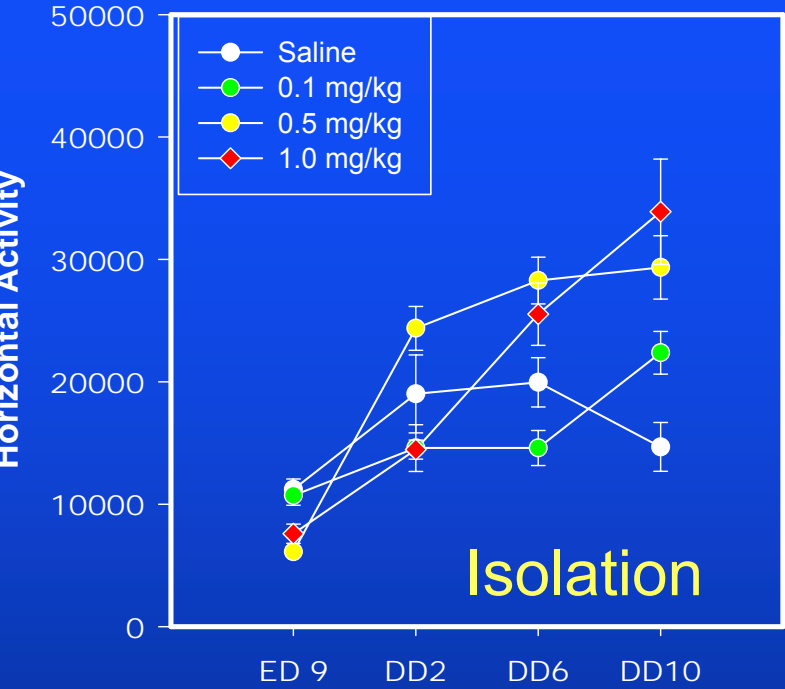
Combined



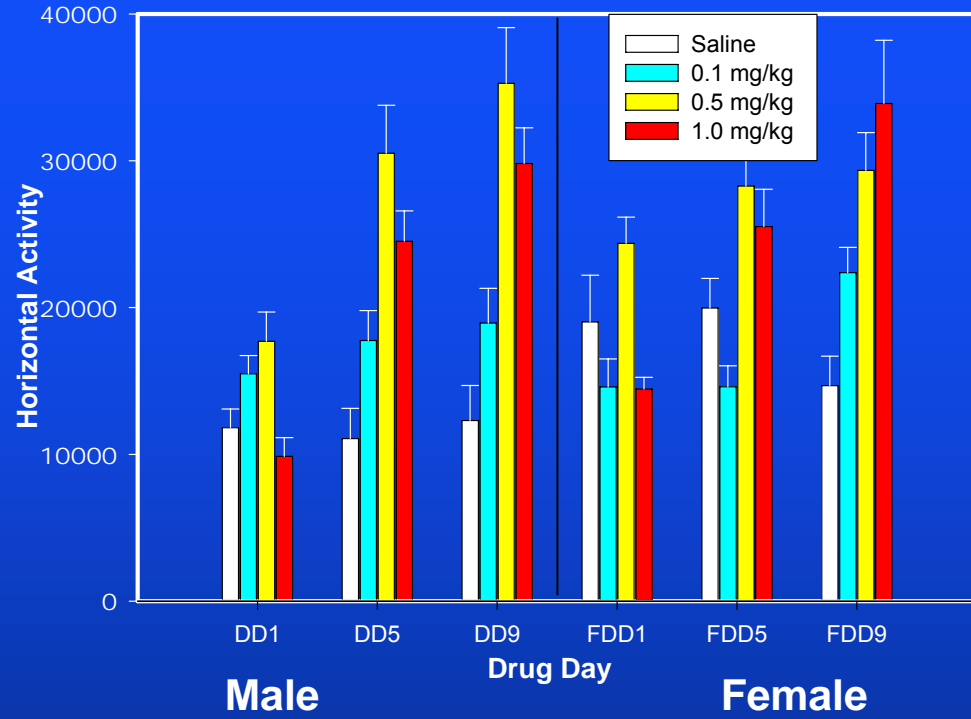
# Effects of Nicotine in Adolescent Males



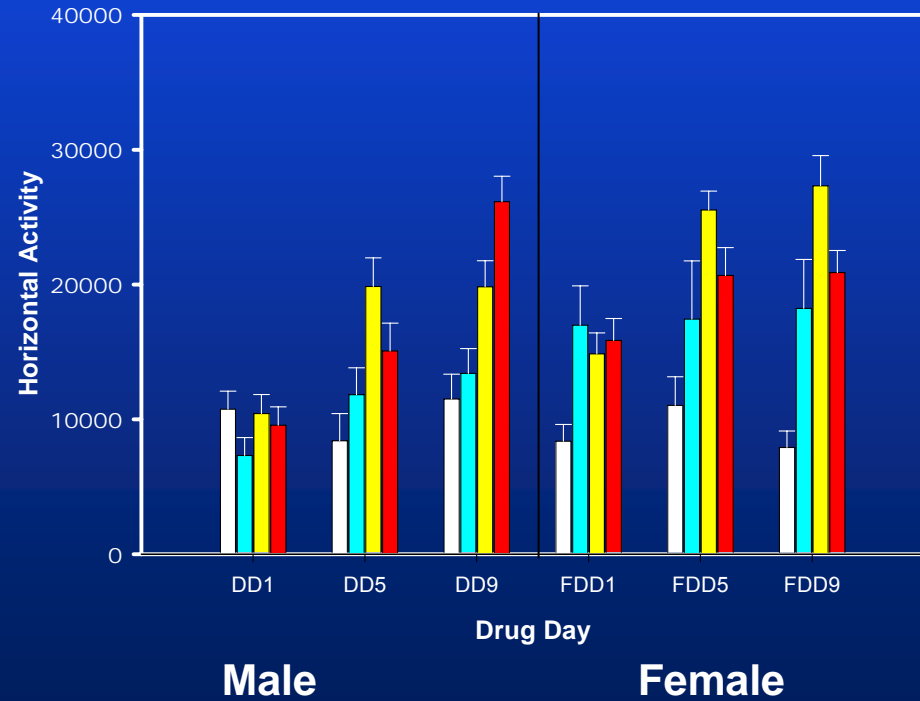
# Effects of Nicotine in Adolescent Females



# Isolation



# Enrichment



# Nicotine and Environment: Results

- Greatest activity in isolated animals that received nicotine regardless of sex
- In male rats, dose-response curve shifted downwards with enrichment vs. isolation
- In female rats, dose-response curve shifted to the left for enrichment vs. isolation

# Nicotine and Environment: Clinical Implications

- For males raised in enriched environments, low nicotine cigarettes may be less rewarding
- For females raised in enrichment environments, low nicotine cigarettes may be more rewarding
- What is the “optimal” amount of enrichment to minimize rewarding effects of addictive drugs?

# Nicotine and Withdrawal

- Scientific Question
  - Are there age and/or sex differences in nicotine withdrawal?
- Clinical Question
  - Are there age and/or sex differences in withdrawal symptoms?

# Nicotine and Withdrawal

- **Subjects**

- 96 male and female SD rats, about 50 days old

- **Design**

- 2 (m or f) X 2 (saline or nicotine) X 2 (stressful or non-stressful environment) X 5 (phase: baseline, nicotine, withdrawal days 1, 2, 3); n = 12 per cell

- **Nicotine administration**

- via SC osmotic mini-pump for 7 days

- **Dependent Measures**

- Observations of withdrawal behaviors (e.g., body shakes, excessive grooming) for 20 minute periods during each phase
- Locomotion in Open Field
- Body Weight
- Estrus measurements



# Nicotine and Withdrawal: Preliminary Results

- All rats displayed withdrawal behaviors
- Male rats display more withdrawal behaviors in a mildly stressful environment than in a non-stressful environment
- Adult females displayed somewhat more withdrawal than adult males in non-stressful environment
- There are genetic-based sex differences in withdrawal behaviors that are evident in adolescent rats

# Nicotine and Withdrawal: Clinical Implications

- Withdrawal symptoms may last longer in certain groups populations compared with others
- Stress management should be incorporated into smoking cessations programs



# Some of our conclusions

- Nicotine alters body weight, attention, stress, and pain
- Body weight effects:  $M < F$ ;  $m > f$
- Anxiety:  $M = F$ ;  $m$  (anxiolytic)  $>$   $f$  (anxiogenic)
- Attention:  $F > M$
- Pain:  $M > F$ ;  $m = f$
- Enrichment:  $m$  dose-response  $\downarrow$   
 $f$  dose-response  $\leftarrow$
- Withdrawal:  $F > M$ ;  $f > m$   
(depending on environment and genetics)

# Current Nicotine Research

- Strain/genetic differences
  - Focus on differences in withdrawal



- Environmental enrichment & nicotine differences
  - On measures of food consumption, body weight, & exercise



- Proteomics
  - In brain regions relevant to different actions of nicotine



# Tobacco/Nicotine: Current and Future Directions for Animal work

- Nicotine and immunopharmacology
- Genomics of individual differences
  - PK and PD
- Interactions with other drugs
- Interactions with environment
- Nicotine as a treatment for medical conditions  
(psychological, neurological, and pathophysiological)

# **Animal models and behavioral medicine: Current and future directions**

- CVD, cancers, diabetes, GI disorders
- Brain injuries, pain
- Sleep disorder, sexual dysfunction, stress disorders
- Substance abuse and interaction with physical and mental health
- Many topics in behavioral medicine deserve parallel, translational research

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