Published in final edited form as:

Alcohol. 2010 February ; 44(1): 57–66. doi:10.1016/j.alcohol.2009.09.035.

Alcohol during adolescence selectively alters immediate and long-term behavior and neurochemistry

Antoniette M. Maldonado-Devincci, M.A.¹, Kimberly A. Badanich, Ph.D.³, and Cheryl L. Kirstein, Ph.D.^{1,2,*}

¹Department of Psychology, Cognitive and Neurosciences, University of South Florida 4202 E. Fowler Avenue Tampa, FL 33620

²Department of Molecular Pharmacology & Physiology, University of South Florida College of Medicine, Tampa, FL 33612

³Center for Drug and Alcohol Programs, Medical University of South Carolina, Charleston, SC 29425

Abstract

Alcohol use increases across adolescence and is a concern in the United States. In humans, males and females consume different amounts of alcohol depending on the age of initiation and the long-term consequences of early ethanol consumption are not readily understood. The purpose of our work is to better understand the immediate and long-term impact of ethanol exposure during adolescence and the effects it can have on behavior and dopaminergic responsivity. We have assessed sex differences in voluntary ethanol consumption during adolescence and adulthood and the influence of binge ethanol exposure during adolescence. We have observed that males are sensitive to passive social influences that mediate voluntary ethanol consumption and early ethanol exposure induces long-term changes in responsivity to ethanol in adulthood. Exposure to moderate doses of ethanol during adolescence produced alterations in dopamine (DA) in the nucleus accumbens septi (NAcc) during adolescence and later in adulthood. Taken together, all of these data indicate the adolescent brain is sensitive to the impact of early ethanol exposure during this critical developmental period.

Keywords

Adolescence; alcohol; sex differences; voluntary alcohol intake; social interaction; dopamine; neurochemistry

© 2009 Elsevier Inc. All rights reserved.

*To whom correspondence should be addressed: Cheryl L. Kirstein, Ph.D., Psychology Department-PCD 4118G, University of South Florida, 4202 East Fowler Avenue, Tampa, Florida 33620, Phone: (813) 974-9626, Fax: (813) 974-4617, kirstein@cas.usf.edu.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

Introduction

Initiation of alcohol use during adolescence is high in both males and females, with rates of use starting as low as 3.9% among adolescents aged 12–13 and increasing rapidly across adolescence with rates as high as 51.6% among individuals aged 18–20 (Substance Abuse and Mental Health Services Administration, 2007). Underage drinking is slightly higher in males than females (Substance Abuse and Mental Health Services Administration, 2007). Our work focuses on the hypothesis that early initiation of alcohol use may be a predictor of continued use into adulthood and those that begin use of alcohol at an early age may also be at risk for developing an alcohol abuse disorder in adulthood (Grant et al., 2001; Hasin and Glick, 1992). Early exposure to ethanol may predispose the brain to greater vulnerability to subsequent ethanol exposure both behaviorally and neurochemically.

Rodent Model of Alcohol Exposure during Adolescence

In rodent models, the time frame encompassing adolescence has yet to reach consensus. Adolescence in the rodent has been defined using various factors indicative of developmental transitions in human adolescents. These factors include changes in behavioral patterns, hormonal patterns, and/or primary sexual characteristics. In a comprehensive review, Spear (2000) argues that adolescence cannot be defined based solely on the characteristics associated with puberty and sexual maturation, but rather, that adolescence is a period of ‘soft events’ that should be viewed as a time of transitions that cannot be clearly delineated. Using the appearance of growth spurt, pruning of excitatory synapses as well as unique behavioral transitions in the rat (e.g., increased peer interaction, play and exploratory behavior in the wild), Spear (2000) defined adolescence broadly from postnatal day (PND) 28 to PND 42, with the acknowledgement that in males some traits may appear as late as PND 55. Other reviews have suggested similar broad classifications defined by the appearance of mature hormonal cycling (PND 28–30) and the subsequent appearance of reproductive maturity which can occur as late as PND 60 in males (Ojeda and Urbanski, 1994; Smith, 2003). Within this broad range, some researchers have identified the categories of early- (PND 21–34), mid- (PND 34–46) and late- (PND 46–59) adolescence centered around the appearance of puberty (between PND 33–44) and encompassed by weaning on PND 21 and reproductive maturity on PND 59 (Tirelli et al., 2003). The beginning of preadolescence in the rat, in terms of sexual maturation begins at approximately PND 20 for females and PND 30 for males (Odell, 1990) with significant changes continuing between PND 28–55 (Ojeda & Urbanski, 1994).

Ethanol has been shown to affect multiple neurotransmitter systems during early development as well as adulthood. Most studies to date have not focused on adolescent animals but have been conducted during the early postnatal period (Druse, 1992; Light, Serbus & Santiago, 1989; Martin, Savage & Swartzwelder, 1992; Savage et al., 1991) or in older, mature animals (see Hunt, 1985, for review). Therefore it is our aim to investigate the behavioral effects of alcohol during adolescence and to determine how alcohol’s effects on mesolimbic DA within adolescence is critical in addressing the long-term effects of early exposure to alcohol. For these reasons our work has examined the effects of alcohol exposure during the adolescent transition period from PND 30–50.

Social Interaction

One of the behavioral aspects that guides some of the recent work we have conducted includes a preclinical approach to understanding environmental factors, specifically the influence of social interaction and its impact on passive social modeling behavior, which can influence early alcohol-seeking behavior in adolescent male and female rats. Social influences, including passive social influences, such as social modeling, contribute to future alcohol use in adolescent humans (Graham, Marks and Hansen, 1991). Previous studies have shown the ability of alcohol to facilitate social interactions as an important factor in adolescent drinking behaviors (Beck, Thombs and Summons, 1993; Beck and Treiman, 1996; Smith, Goldman, Greenbaum and Christiansen, 1995; Thombs, Beck, Mahoney, Bromley and Bezon, 1994).

In recent work, we have employed the demonstrator-observer paradigm developed by Strupp and Levitsky (1984). The paradigm is based on the interaction between two animals; one serving as a demonstrator, which serves as part of the experimental paradigm and the other as an observer, which serves as the experimental animal in which changes in behavior are assessed. Social behaviors peak during adolescence, and adolescent rats engage in distinctive forms of social behavior, including play behavior, social investigation and social contact (Panksepp, 1981; Pellis, Field, Smith and Pellis, 1997; Pellis and Pellis, 1997; Varlinskaya, Spear and Spear, 2001, Varlinskaya and Spear, 2002). It has been suggested that social play represents a separate category of behavior that is different from adult typical social, sexual and aggressive types of behaviors (for review see Vanderschuren et al., 1997). While play behavior peaks during adolescence, as adults mature their behavior becomes more aggressive (Meaney and Stewart, 1981). These data demonstrate that the peak in social behavior is present during the adolescent period of development, and not in younger or older animals. Therefore, assessing the influence of social behaviors on passive social learning specifically during adolescence is appropriate.

Acute ethanol increases social interaction in adolescent rats (Varlinskaya et al., 2001), and produces social inhibition in adult rats (Varlinskaya and Spear, 2006). Given the increased social interaction observed in adolescent rats administered ethanol, the demonstrator-observer model has been applied to assess preference for alcohol in adolescent rodents after social interaction with a familiar alcohol-intoxicated peer (e.g. Fernandez-Vidal and Molina, 2004; Hunt, Holloway and Scordalakes, 2001). One experiment was designed to extend on the findings of Hunt and colleagues, in which they observed increased ethanol intake in adolescent rats following social interaction with a familiar alcohol-intoxicated peer (Hunt et al., 2001). The present work assessed the effects of social familiarity during social interaction on voluntary ethanol intake using a two-bottle choice test with a choice between a sweetened ethanol solution and water in male and female adolescent rats (Maldonado, Finkbeiner and Kirstein, 2008). Consistent with previous work in rodents (Hunt et al., 2001), we found adolescent males and females that socially interacted with an alcohol-intoxicated familiar peer voluntarily consumed significantly more sweetened ethanol relative to those that socially interacted with an alcohol-free peer (Maldonado et al., 2008a). In contrast, adolescent males that socially interacted with an unfamiliar alcohol-intoxicated peer consumed significantly less sweetened ethanol relative to adolescent males that socially

interacted with an unfamiliar alcohol-free peer (Maldonado et al., 2008a). Adolescent females that socially interacted with an unfamiliar alcohol-intoxicated peer showed similar increases in voluntary sweetened ethanol intake relative to those that socially interacted with a familiar alcohol-intoxicated peer (Maldonado et al., 2008a). It is interesting to note the striking sex differences observed between males and females when the paradigm was altered by familiarity of the peer.

The experiments described above indicate exposure to an alcohol-intoxicated peer altered ethanol consumption after just one pairing of social interaction with an alcohol-intoxicated peer. In the experiment described below (Figure 1), preliminary data are presented to further investigate the impact of repeated pairings of environmental cues paired with social interaction on preference for the associated environment in alcohol-naïve adolescent observer rats. In this experiment (Figure 1), we aimed to expand on the findings of Fernandez-Vidal and Molina (2004), in which they found that adolescent rats that repeatedly socially interacted with an alcohol-intoxicated peer showed a change in preference for an alcohol odor as compared to an appetitive banana odor. It is important to note in this social interaction experiment, the observer was never orally administered water or ethanol, therefore the observer rats were in a drug-free state and we assessed the passive social influence of social interaction with an alcohol-intoxicated peer on changes in behavior in the ethanol-naïve adolescent observer. Specifically, we assessed the impact of repeated pairings of social interaction with an alcohol-intoxicated peer paired with environmental cues on changes in preference for the chamber paired with social interaction with an alcohol-intoxicated peer in adolescent male and female rats. Our paradigm followed the same age and number of pairings of social interaction with an alcohol-intoxicated peer as that used in the Fernandez-Vidal and Molina (2004) study. In the present work, rats were given a 15-minute baseline conditioning assessment on PND 30 in which rats had free access to both chambers to assess baseline preferences for each chamber. The chamber the adolescent observer rat spent the least amount of time in was paired with social interaction with an alcohol-intoxicated peer. Control rats socially interacted with the same alcohol-free peer in both chambers. Conditioning occurred over a period of four days from PND 31–34. Specifically, in the morning, all rats underwent social isolation in a holding cage and water administration to the demonstrator after the initial 30 minutes of social isolation followed by an additional 30 minutes of social isolation. Rats were then given the opportunity to socially interact with an alcohol-free peer in the initially most-preferred chamber for 30 minutes. Four hours later, the same paradigm was repeated, however the same demonstrator was orally administered ethanol (0.5 or 1.5 g/kg) or water in control rats and the dyad were confined to the alternate chamber for 30 minutes. Each adolescent observer socially interacted with the same adolescent demonstrator for all social interaction sessions. However, the demonstrator was sober in the morning sessions of social interaction and intoxicated in the afternoon sessions of social interaction. Control rats socially interacted with the same sober peer for all social interaction sessions. For each dose tested in which the observer socially interacted with an alcohol-intoxicated or non-intoxicated demonstrator, separate rats were used, for a total of three groups of observer-demonstrator dyads (control, 0.5 and 1.5 g/kg). This cycle was repeated three additional times for a total of four pairings with an alcohol-free peer and four pairings with an alcohol-intoxicated peer (or eight

pairings with an alcohol-free peer for control rats). Each observer socially interacted with the same demonstrator for all conditioning sessions. On the final day, PND 35, all rats underwent a post-conditioning test to assess changes in time spent in the designated paired chamber (Maldonado and Kirstein, unpublished data).

Note that adolescent observers were never administered water or ethanol in this experiment, but were given the opportunity to socially interact with an alcohol-intoxicated demonstrator to assess changes in behavior. As depicted in Figure 2, adolescent ethanol naïve observer male rats that socially interacted with an alcohol-intoxicated peer administered the highest dose (1.5 g/kg) showed a significant increase in time spent in the chamber paired with the alcohol-intoxicated peer at the post-conditioning test relative to the preconditioning baseline assessment as compared to all other groups. There were no other significant differences observed in adolescent ethanol naïve male observers that socially interacted with an alcohol-intoxicated peer administered the lower ethanol dose (0.5 g/kg) relative to their controls. Nor were there changes in preference for the environment paired with social interaction with an alcohol-intoxicated demonstrator at either dose in ethanol-naïve female observers relative to their controls (Maldonado and Kirstein, unpublished data). Together, these data indicate adolescent male rats are especially sensitive to environmental conditions surrounding the influence of the peer on changes in behavior following social interaction with an alcohol-intoxicated peer.

Social play in adolescent animals has been suggested to be rewarding using the conditioned place preference (CPP) paradigm (Calcagnetti & Schechter, 1992). Consistent with Calcagnetti and Schechter (1992), there were no significant sex differences in time spent in the initially least preferred side after conditioning in control animals in the social interaction conditioning experiment discussed above (Figures 1 and 2). Extending upon the work of Calcagnetti and Schechter (1992), using the conditioned place preference paradigm in adolescent animals, in conjunction with the effects of ethanol on facilitating social interaction, adolescent male rats appear to be the most sensitive to the effects of cues paired with social interaction with an alcohol-intoxicated peer. Overall, the present results suggest adolescent males are particularly sensitive to environmental conditions when socially interacting with an alcohol-intoxicated peer, which may alter subsequent responding for alcohol in social settings.

The ability of alcohol to facilitate social interactions plays a key role in adolescent drinking behaviors (Beck et al., 1993; Beck and Treiman, 1996; Smith et al., 1995). Additionally, one of the most important factors implicated in alcohol use in high school students is its' effects on social facilitation (Beck & Treiman, 1996; Thombs et al., 1994). Passive social influences have been shown to affect future alcohol use in adolescent humans (Graham et al., 1991) and the results obtained from the present experiments and previous preclinical work (Fernandez-Vidal & Molina, 2004; Hunt et al., 2001) confirms this notion. More work should be conducted to identify the specific mechanisms that may mediate these results and how they may alter adolescent alcohol-seeking behavior.

Voluntary Ethanol Consumption

The method of initial exposure to ethanol can alter future drinking patterns in rats (Files et al., 1997). Ethanol consumption during adolescence appears to have no effect on subsequent ethanol intake in adulthood (Slawecki and Betancourt, 2002; Tolliver and Samson, 1991; Vetter et al., 2007). Adolescent rats voluntarily consume more ethanol relative to adults (Truxell et al., 2007) when animals were allowed voluntary access to ethanol under continuous access conditions (Brunell and Spear, 2005; Doremus et al., 2005; Vetter et al., 2007). The same effect was observed using a modified sucrose-substitution paradigm under limited access conditions (Maldonado, Finkbeiner, Alipour and Kirstein, 2008).

Importantly, some of our most recent work has examined some of the sex-related differences that may accompany the age-related differences in voluntary ethanol consumption under limited access conditions. In the most common paradigm we employ in our experiments, rats are given daily 30 minutes access to sweetened ethanol solutions using sucrose or saccharin. Rats are initially placed in a holding cage for 30–60 minutes to allow them to acclimate to the behavioral testing room and subsequently given access to ethanol and water for 30 minutes in rats that are neither food, nor water deprived. We have observed interesting sex and age-related differences in voluntary ethanol consumption under these limited access conditions. As described recently, adolescent male rats consume significantly more ethanol than adult male rats at higher sweetened ethanol concentrations (10% and 20% v/v ethanol) (Maldonado et al., 2008b). In contrast, adult female rats consume significantly more ethanol than adolescent female rats at lower sweetened ethanol concentrations (5% and 10%). Consistent with other work, we have observed increased voluntary sweetened ethanol intake in females as compared to males, when voluntary sweetened ethanol intake was equated for body weight. These data highlight the importance of examining sex differences when conducting developmental experiments. It appears early during adolescence there are no differences in voluntary sweetened ethanol consumption (Maldonado and Kirstein, unpublished data), however sex differences in voluntary ethanol consumption are observed in adulthood (Almeida et al., 1998; Blanchard et al., 1993; Juarez and Barrios de Tomasi, 1999; Lancaster et al., 1996). Additionally, across development from adolescence into young adulthood, there appears to be a natural decrease in voluntary ethanol consumption (Vetter et al., 2007). Given that sex differences emerge in adulthood, early exposure to ethanol during adolescence could alter the normal pattern of ethanol consumption in adulthood.

Interestingly, when using a binge model of adolescent ethanol exposure and assessing subsequent sweetened ethanol intake under limited access conditions in young adulthood in male and female rats, we have observed dramatic increases in voluntary ethanol intake in rats that were treated with high doses of ethanol during adolescence relative to those that were treated with water during adolescence (Maldonado et al., 2009). Male and female rats were exposed to ethanol (1.5, 3.0 or 5.0 g/kg/intragastric intubation) or water across adolescence in four-day intervals (PND 28–31, PND 35–38, and PND 43–45) with three days between each administration in which animals were left undisturbed (PND 32–34 and PND 39–41). Immediately following ethanol pretreatment during adolescence, all rats underwent fourteen days of abstinence in which animals were left undisturbed (PND 46–59).

Finally, voluntary sweetened ethanol (0.5% saccharin/ 10% ethanol) intake was assessed in young adulthood (PND 60–69). According to preliminary data, when ethanol intake was equated for body weight, females consumed significantly more ethanol than males regardless of pretreatment with ethanol or water. Pretreatment with any dose of ethanol enhanced this effect. However, when we equated for the innate sex differences in voluntary ethanol consumption observed in adulthood, it appears males were more vulnerable to the subsequent increase in voluntary ethanol consumption in adulthood. These data highlight the importance of early ethanol history and the relative impact it has on later ethanol consumption in adulthood when assessing vulnerability to subsequent ethanol-seeking and consumption behaviors.

Long-term effects of adolescent ethanol exposure

Novelty preference has been extensively examined in the literature as a measure of individual differences and its influence in responsivity to drugs of abuse. We have established a paradigm that allows us to assess novelty preference in adolescent and adult animals using a single novel object in a circular open field. Using this paradigm, we have readily demonstrated adolescent rats display greater levels of novelty preference relative to adult rats (Stansfield and Kirstein, 2006). Interestingly, using this paradigm, we found chronic pretreatment (PND 30–50) with a moderate dose of ethanol during adolescence (1.0 g/kg/ip) significantly increased novelty seeking behaviors manifested as a greater frequency to approach a novel object in the center of the open field (Stansfield and Kirstein, 2007). Given that novelty preference and risk-taking behaviors have repeatedly been associated with an increased propensity to drug self-administration and greater drug intake (Bevins et al., 1997), these data support the hypothesis that adolescent rats chronically exposed to ethanol during adolescence may be at even greater risk to greater ethanol intake in adulthood.

Differential biphasic effects of ethanol have been demonstrated for ethanol-induced motor activity. Ethanol-induced locomotor activity has been shown to be increased among adolescent male and female alcohol-preferring P rats (Rodd et al., 2004). Specifically, in response to an acute dose of 0.50 or 0.75 g/kg ethanol, adolescent P female rats demonstrated increased activity counts relative to animals administered saline or 0.25 g/kg ethanol. Additionally, adolescent P males exhibited elevated activity counts when administered 0.25, 0.50, or 0.75 g/kg ethanol relative to saline controls. In animals of both sexes activity counts were significantly decreased at 1.50 g/kg ethanol. Because of the heightened sensitivity to ethanol among adolescent P females, the present study used out-bred female rats to assess the effects of chronic ethanol administration (PND 34–46) during adolescence to assess the dose-dependent effects of ethanol on locomotor activity to 0.75 and 1.50 g/kg/ip/b.i.d. ethanol (Maldonado and Kirstein, unpublished data). Rats underwent abstinence from PND 47 to PND 59. Rats were then challenged with ethanol (0.75 or 1.50 g/kg/ip) or saline on PND 60.

The results of this study indicated significant long-term alterations in behavior among adolescent female rats that were repeatedly exposed to ethanol during this period of development. Adolescent females developed tolerance to the high dose of ethanol (1.50

g/kg) during repeated exposure as evidenced by an initial acute suppression in locomotor activity in ethanol-treated (PND 35) rats followed by the development of similar levels of locomotor activity between ethanol and saline-treated (PND 46) rats. However, this pattern did not persist into adulthood following a washout period between adolescence and adulthood, as ethanol-treated rats showed decreased locomotor activity in young adulthood when challenged with the same dose of ethanol on PND 60. It is evident that the low dose of ethanol produced a facilitation of locomotor activity when tested after repeated ethanol exposure during adolescence, as rats treated with the lower (0.75 g/kg) dose of ethanol showed increased locomotor activity (PND 46) following repeated ethanol treatment during adolescence relative to saline-treated rats. This pattern persisted into young adulthood when animals were challenged with the same moderate dose of ethanol on PND 60 (Maldonado and Kirstein, unpublished data). Interestingly, behavioral sensitization has been observed in Sprague Dawley rats to a lower dose of ethanol when animals were separated into high and low responders to novelty (Hoshaw & Lewis 2001). These results again demonstrate repeated alcohol exposure during adolescence results in persistent alterations in behavior that last into young adulthood.

Adolescent exposure to ethanol also tended to alter responsivity in a conditioned place preference paradigm in young adulthood. Adolescent male rats were chronically exposed to saline or ethanol (0.75 g/kg/ip) during adolescence (PND 30–50) followed by a 14-day washout period and were conditioned with saline or ethanol (0.75 g/kg/ip) in young adulthood in a two-chambered apparatus (Maldonado, Badanich and Kirstein, unpublished data). In young adulthood (PND 65–70), rats were conditioned with saline or ethanol. Control rats had saline paired with each environment for a total of eight saline conditioning trials. For rats conditioned with ethanol in adulthood, four pairings with saline and four pairings with ethanol occurred. As shown in Figure 3, adolescent rats that were pretreated with ethanol and conditioned with ethanol in young adulthood spent significantly more time in the ethanol-paired chamber relative to those adolescent rats that were pretreated with saline and conditioned with ethanol in young adulthood. Additionally, adolescent rats that were treated with ethanol and subsequently conditioned with ethanol in young adulthood demonstrated a greater amount of time spent in the ethanol-paired chamber relative to animals pretreated with ethanol and conditioned with saline during young adulthood (Figure 3; Maldonado, Badanich, and Kirstein, unpublished data). These data indicate animals exposed to ethanol during adolescence and re-exposed to ethanol in adulthood showed greater ethanol-seeking behavior manifested as increased time spent in the drug-paired chamber relative to animals exposed to ethanol for the first time in adulthood.

Taken together, all of these data indicate exposure to ethanol during adolescence does indeed alter multiple behaviors relative to ethanol-naïve animals. These data support the hypothesis that the adolescent brain is sensitive to insult induced by ethanol exposure during adolescence and these changes are manifested behaviorally as increased vulnerability to the effects of ethanol in young adulthood. However, there are several areas in the brain that may be uniquely susceptible to the long-term effects of early ethanol exposure, including the mesolimbic dopaminergic system, which is one of the key systems that mediate the behavioral changes we have observed in our work. We focus our neurochemical studies on

the mesolimbic dopaminergic system given its involvement in reward, attention, novelty preference and salience.

Neurochemistry

Alcohol consumption during adolescence is frequent and often involves heavy drinking patterns such as binge drinking (Johnston, O'Malley, Bachman and Schulenberg, 2006). High rates of alcohol consumption have been reported for human (Bates and Labouvie, 1997) and rodent adolescents in comparison to adults (Doremus et al., 2005). Normal development of neuronal pathways may be disturbed by alcohol consumption during adolescence, consequently altering subsequent adult brain functioning. Therefore, it is important to understand how the adolescent brain develops and also the impact of early alcohol exposure on adult behavior and brain functioning.

Alcohol abuse research in rodents has focused on one particular pathway in the brain, the mesolimbic dopamine (DA) pathway that originates in the ventral tegmental area (VTA) and projects to the nucleus accumbens septi (NAcc; Dahlstrom and Fuxe, 1964). Due to the fact that ethanol increases DA in the NAcc as measured by *in vivo* microdialysis (Yoshimoto et al, 1992), examining ethanol's impact on extracellular DA levels in adolescents would help determine whether DA mediates adolescent vulnerability to high ethanol consumption rates and alcohol dependency.

Our early work showed preadolescent rats (PND 25) had increased NAcc DA levels following acute or repeated ethanol injections (1.0 g/kg/ip) relative to saline injected controls with repeated ethanol inducing a leftward shift in peak DA levels (Philpot and Kirstein, 1998). Therefore, ethanol-induced DA levels could be reliably obtained in the young rat. The ability of ethanol to induce DA associated expectancy effects was also measured. Preadolescent rats pretreated with ethanol but only given a saline injection during DA sampling exhibited increased NAcc DA levels as well (Philpot and Kirstein, 1998). These data suggest DA levels increased in anticipation or expectancy of ethanol administration.

Recently, both conventional *in vivo* microdialysis and quantitative *in vivo* microdialysis under transient conditions were performed to determine whether age-related differences in basal DA exist in the NAcc. Preadolescent (PND 25), early adolescent (PND 35), late adolescent (PND 45) and young adult (PND 60) rats were implanted with cannula and microdialysis probes into the NAcc. Using the conventional approach, artificial cerebrospinal fluid was perfused through the probe and DA was recovered (Philpot and Kirstein, 2004). Using the quantitative approach (adopted from Olson & Justice, 1993) differing concentrations of DA were perfused and the dialysate analyzed to determine levels based on the concentration put in and the concentration recovered (Badanich, Adler and Kirstein, 2006). The concentration that reached steady state (DA in = DA out) was considered the basal level. Both approaches yielded similar age results, with late adolescent animals (PND 45) having significantly higher basal DA levels than adults, but with the conventional method (Philpot and Kirstein, 2004), dialysate DA levels obtained in adolescents were higher than those obtained using the quantitative method (Figure 4; Badanich et al., 2006). Additionally, the quantitative approach revealed lower basal DA

levels for early adolescent (PND 35) rats. It should be noted the conventional method reported dialysate DA levels that were not corrected for probe recovery. These results indicate ontogenetic differences in basal DA. Furthermore, different findings for PND 35 rats between the two methods suggest that reuptake and/or metabolic differences for early adolescent rats given that the quantitative method indirectly assesses group differences in reuptake and transporter functioning. These data characterized basal DA levels in naive adolescent rats and suggest basal inherent differences in DA during development may likely impact neurochemical reactivity to early life alcohol exposure and produce long-term consequences in neuronal functioning in adulthood.

Early alcohol use is highly associated with more severe alcohol dependency as an adult (Grant and Dawson, 1997) and mesolimbic DA mediate the rewarding properties of alcohol in adult rodents (Engleman et al., 2000; Ericson, Blomqvist, Engel and Soderpalm, 1998; Gonzales, Job and Doyon, 2004; Katner and Weiss, 2001; Weiss, Lorang, Bloom and Koob, 1993; Weiss et al., 1996). Given these data, it was hypothesized that adult rats pretreated with ethanol during adolescence would have different basal DA concentrations than saline-treated controls. Adolescent rats were injected daily from PND 30–50 with either 0.75 g/kg/ip ethanol or saline followed by an ethanol-abstinent period from PND 51–65. Changes in extracellular DA levels in the NAcc were quantified on PND 70. Extracellular DA levels were greater in rats chronically treated with ethanol during adolescence in comparison to saline-exposed controls (Figure 5; Badanich, Maldonado and Kirstein, 2007). These data support similar reports in the adult literature (Weiss et al., 1996); however, a recent adolescent alcohol report (Sahr, Thielen, Lumeng, Li and McBride, 2004) suggested there were no differences in DA for rats selectively bred to drink alcohol during adolescence. Differences between the previous adolescent report and the present study may certainly be attributable to differences in route of administration whereas the previous report incorporated voluntary self-administration and the present study incorporated passive experimenter administration of alcohol injections. Voluntary self-administration of alcohol produces different neurochemical responses than involuntary or passive administration of alcohol (Ericson et al., 1998; Weiss et al., 1993; Imperato and Di Chiara, 1986; Kiianmaa, Nurmi, Nykanen and Sinclair, 1995; Yoshimoto, McBride, Lumen and Li, 1992). DA results may have differed had rats in the present study been allowed to voluntarily drink alcohol during adolescence. Further, differences between the previous report and the present study may be attributable to genetic differences between rodent strains particularly because rats used in the previous report were selectively bred for high ethanol consumption while rats in the present study were out-bred rats.

These data provided insight into how the adolescent brain responds to ethanol insult during adolescence and that these effects persist into adulthood. These data contribute to an understanding of how the human adolescent brain may develop and also how alcohol use during adolescence effects normal brain development. Clinical and experimental research should continue to focus on the development of the adolescent brain so to better understand how to treat adolescent substance use/abuse. Future research should investigate whether elevated basal DA impacts ethanol-seeking behavior differently in adolescent and adult rats as DA has been shown to mediate drug-seeking behavior in adult rodents.

Summary and Future Directions

We have assessed sex-differences in voluntary ethanol consumption during adolescence and adulthood and the influence of binge ethanol exposure during adolescence. We have demonstrated that males are sensitive to passive social influences that mediate voluntary ethanol consumption and that early ethanol exposure induces long-term changes in responsivity to ethanol in adulthood. Neurochemically, we have demonstrated exposure to moderate doses of ethanol during adolescence produced alterations in DA during adolescence and later in adulthood. All of these data indicate the adolescent brain is sensitive to the impact of early ethanol exposure during this critical developmental period. It is important to note that these effects may be limited by the age of ethanol exposure and dose administered. Therefore, it is important to assess if these effects observed in our work are specific to exposure during adolescence or if similar effects would be observed if exposure was initiated in adulthood.

Our work has primarily focused on characterizing alcohol consumption patterns, socialization with intoxicated peers and alcohol-induced changes in NAcc DA levels of adolescent and adult rats. Many questions regarding sensitivity to alcohol during adolescence still need to be addressed including how long do these age-related changes in behavioral and dopaminergic responsivity persist? Are changes in sensitivity to alcohol throughout development dependent on dose or exposure pattern? Does early alcohol exposure induce cognitive deficits that persist into adulthood such as changes in behavioral inhibition, decision-making, memory and attending to salient stimuli. Future experiments should utilize behavioral measures dependent on cortical activity such as the effects of chronic alcohol exposure on working memory, set-shifting, impulsivity and reversal learning. It would be interesting to explore whether alcohol use and/or dependency during adolescence exacerbates deficits in performance on cognitive tasks such as slowing the acquisition rate or by increasing the perseveration rate on a cortical-dependent task. While recently we have begun to examine sex differences in the impact of early ethanol exposure on behavior, future work should be extended to examine sex differences in NAcc DA following early ethanol exposure. Given that research has been conducted on the interaction of glutamate and dopamine following chronic exposure to ethanol exclusively in adult rodents (Kapasova and Szumlinski, 2008; Szumlinski et al, 2007; Piepponen et al, 2002), it would be interesting to evaluate these same neurochemical interactions in male and female adolescent rats and to determine if any alterations persist into adulthood. It is plausible that both DA and glutamate interact to induce long-lasting changes in the mesolimbic pathway and cortical areas since ethanol has been shown to alter synaptic efficacy of both glutamatergic and dopaminergic pathways (Carpenter-Hyland and Chandler, 2007; Yim and Gonzales, 2000; Smith and Weiss et al, 1999; Philpot et al, 1998; Badanich et al, 2007). Taking these approaches will allow us to determine underlying neurochemical mechanisms mediating changes in sensitivity to alcohol consumption and other alcohol-related behaviors.

Acknowledgments

This work was funded by NIAAA AA11820, the University of South Florida Neuroscience Health Program, the University of South Florida College of Arts and Sciences, the Department of Psychology and the University of South Florida Honors College.

References

- Almeida OFX, Shoaib M, Deicke J, Fischer D, Darwish MH, Patchev VK. Gender differences in ethanol preference and ingestion in rats: the role of the gonadal steroid environment. *J Clin Invest.* 1998; 101(12):2677–2685. [PubMed: 9637701]
- Badanich KA, Adler KJ, Kirstein CL. Adolescents differ from adults in cocaine conditioned place preference and cocaine-induced dopamine in the nucleus accumbens septi. *Eur J Pharmacol.* 2006; 550:95–106. [PubMed: 17011546]
- Badanich KA, Maldonado AM, Kirstein CL. Chronic ethanol exposure during adolescence increases basal dopamine in the nucleus accumbens septi during adulthood. *Alcohol Clin Exp Res.* 2007; 31(5):895–900. [PubMed: 17391340]
- Bates ME, Labouvie EW. Adolescent risk factors and the prediction of persistent alcohol and drug use into adulthood. *Alcohol Clin Exp Res.* 1997; 21(5):944–950. [PubMed: 9267549]
- Beck KH, Thombs DL, Summons TG. The social context of drinking scales: construct validation and relationship to indicators of abuse in an adolescent population. *Addict Behav.* 1993; 18(2):159–169. [PubMed: 8506787]
- Beck KH, Treiman KA. The relationship of social context of drinking, perceived social norms, and parental influence to various drinking patterns of adolescence. *Addict Behav.* 1996; 21(5):633–634. [PubMed: 8876762]
- Bevins KJ, Bardo MT. Individual differences in response to novelty, amphetamine-induced activity and drug administration in rats. *Behav Pharmacol.* 1997; 8:113–123. [PubMed: 9833007]
- Blanchard BA, Steindorf S, Wang S, Glick SD. Sex differences in ethanol-induced dopamine release in nucleus accumbens and in ethanol consumption in rats. *Alcohol Clin Exp Res.* 1993; 17:968–973. [PubMed: 8279683]
- Brunell SC, Spear LP. Effect of stress on the voluntary intake of a sweetened ethanol solution in pair-housed adolescent and adult rats. *Alcohol Clin Exp Res.* 2005; 29(9):1641–1653. [PubMed: 16205364]
- Calcagnetti DJ, Schechter MD. Place conditioning reveals the rewarding aspect of social interaction in juvenile rats. *Physiol Behav.* 1992; 51(4):667–672. [PubMed: 1594664]
- Carpenter-Hyland EP, Chandler LJ. Adaptive plasticity of NMDA receptors and dendritic spines: implications for enhanced vulnerability of the adolescent brain to alcohol addiction. *Pharmacol Biochem Behav.* 2007; 86(2):200–208. [PubMed: 17291572]
- Dahlstrom A, Fuxe K. Localization of monoamines in the lower brain stem. *Experientia.* 1964; 20(7):398–399. [PubMed: 5856530]
- Doremus TL, Brunell SC, Rajendran P, Spear LP. Factors influencing elevated ethanol consumption in adolescent relative to adult rats. *Alcohol Clin Exp Res.* 2005; 29(10):1796–1808. [PubMed: 16269909]
- Druse, MJ. Effects of in utero ethanol exposure on the development of neurotransmitter systems. In: Miller, MW., editor. *Development of the central nervous system: Effects of alcohol and opiates.* New York: Wiley-Liss; 1992. p. 139–167.
- Engleman EA, McBride WJ, Wilber AA, Shaikh SR, Eha RD, Lumeng L, Li TK, Murphy JM. Reverse microdialysis of a dopamine uptake inhibitor in the nucleus accumbens of alcohol-preferring rats: effects on dialysate dopamine levels and ethanol intake. *Alcohol Clin Exp Res.* 2000; 24(6):795–801. [PubMed: 10888067]
- Ericson M, Blomqvist O, Engel JA, Soderpalm B. Voluntary ethanol intake in the rat and the associated accumbal dopamine overflow are blocked by ventral tegmental mecamylamine. *Eur J Pharmacol.* 1998; 358(3):189–196. [PubMed: 9822883]
- Fernandez-Vidal JM, Molina JC. Socially mediated alcohol preferences in adolescent rats following interaction with an intoxicated peer. *Pharmacol Biochem Behav.* 2004; 79:229–241. [PubMed: 15501298]
- Gonzales RA, Job MO, Doyon WM. The role of mesolimbic dopamine in the development and maintenance of ethanol reinforcement. *Pharmacol Ther.* 2004; 103:121–146. [PubMed: 15369680]
- Graham JW, Marks G, Hansen WB. Social influence processes affecting adolescent substance use. *J Applied Psych.* 1991; 76(2):291–298.

- Grant BF, Dawson DA. Age at onset of alcohol use and its association with DSM-IV alcohol abuse and dependence: results from the National Longitudinal Alcohol Epidemiologic Survey. *J Subst Abuse*. 1997; 9:103–110. [PubMed: 9494942]
- Grant BF, Stinson FS, Harford TC. Age at onset of alcohol use and DSM-IV alcohol abuse and dependence: a 12-year follow-up. *J Subst Abuse*. 2001; 13(4):493–504. [PubMed: 11775078]
- Hasin DS, Glick H. Severity of DSM-III-R alcohol dependence: United States, 1988. *Br J Addict*. 1992; 87(12):1725–1730. [PubMed: 1490086]
- Hosha BA, Lewis MJ. Behavioral sensitization to ethanol in rats: evidence from the Sprague-Dawley strain. *Pharmacol Biochem Behav*. 2001; 68(4):685–690. [PubMed: 11526965]
- Hunt, WA. Alcohol and biological membranes. New York: Guilford Press; 1985.
- Hunt PS, Holloway JL, Scordalakes EM. Social interaction with an intoxicated sibling can result in increased intake of ethanol by periadolescent rats. *Dev Psychobiol*. 2001; 38:101–109. [PubMed: 11223802]
- Imperato A, Di Chiara G. Preferential stimulation of dopamine release in the nucleus accumbens of freely moving rats by ethanol. *J Pharmacol Exp Ther*. 1986; 239(1):219–228. [PubMed: 3761194]
- Johnston, LD.; O'Malley, PM.; Bachmanm, JG.; Schulenberg, JE. NIH Publication No 06–5882. Bethesda, MD: National Institute on Drug Abuse; 2006. Monitoring the future national results on adolescent drug use: Overview of key findings, 2005.
- Juarez J, Barrios de Tomasi E. Sex differences in alcohol drinking patterns during forced and voluntary consumption in rats. *Alcohol*. 1999; 19:15–22. [PubMed: 10487383]
- Kapasova Z, Szumlinski KK. Strain differences in alcohol-induced neurochemical plasticity: a role for accumbens glutamate in alcohol intake. *Alcohol Clin Exp Res*. 2008; 32(4):617–631. [PubMed: 18341649]
- Katner SN, Weiss F. Neurochemical characteristics associated with ethanol preference in selected alcohol-preferring and -nonpreferring rats: a quantitative microdialysis study. *Alcohol Clin Exp Res*. 2001; 25(2):198–205. [PubMed: 11236833]
- Kiianmaa K, Nurmi M, Nykanen I, Sinclair JD. Effect of ethanol on extracellular dopamine in the nucleus accumbens of alcohol-preferring AA and alcohol-avoiding ANA rats. *Pharmacol Biochem Behav*. 1995; 52(1):29–34. [PubMed: 7501675]
- Lancaster FE, Brown TD, Coker KL, Elliot JA, Wren SB. Sex differences in alcohol preference and drinking patterns emerge during the early postpubertal period in Sprague-Dawley rats. *Alcohol Clin Exp Res*. 1996; 20:1043–1049. [PubMed: 8892526]
- Light KE, Serbus DC, Santiago M. Exposure of rats to ethanol from postnatal days 4–8: Alterations of cholinergic neurochemistry in the hippocampus and cerebellum at day 20. *Alcohol Clin Exp Res*. 1989; 13:686–692. [PubMed: 2688469]
- Maldonado AM, Finkbeiner LM, Kirstein CL. Social interaction and partner familiarity differentially alters voluntary ethanol intake in adolescent male and female rats. *Alcohol*. 2008a; 42(8):641–648. [PubMed: 18922665]
- Maldonado AM, Finkbeiner LM, Alipour KK, Kirstein CL. Voluntary ethanol consumption differs in adolescent and adult male rats using a modified sucrose-fading paradigm. *Alcohol Clin Exp Res*. 2008b; 32(9):1574–1582. [PubMed: 18616665]
- Maldonado AM, Levie KE, Alipour KK, Kirstein CL. Voluntary sweetened ethanol intake is increased in young adulthood following binge ethanol treatment during adolescence in male and female rats. *Alcohol Clin Exp Res*. 2009; 33(s1):96A.
- Martin D, Savage DD, Swartzwelder HS. Effects of prenatal ethanol exposure on hippocampal ionotropic-glutamate and kainate receptors. *Alcohol Clin Exp Res*. 1992; 16:816–821. [PubMed: 1382392]
- Meaney MJ, Stewart J. A descriptive study of social development in the rat (*Rattus norvegicus*). *Animal Behav*. 1981; 29:34–45.
- Odell, WD. Sexual maturation in the rat. In: Grumbach, MM.; Sizonenko, PC.; Aubert, ML., editors. Control of the onset of puberty. Baltimore, MD: Williams & Wilkins; 1990. p. 183–210.
- Ojeda, SR.; Urbanski, HF. Puberty in the rat. In: Knobil, E.; Neill, JD., editors. The Physiology of Reproduction. 2. New York: Raven Press; 1994. p. 363–409.

- Olson RJ, Justice JB Jr. Quantitative microdialysis under transient conditions. *Anal Chem.* 1993; 65(8):1017–1022. [PubMed: 8494171]
- Panksepp J. The ontogeny of play in rats. *Dev Psychobiol.* 1981; 14(4):327–332. [PubMed: 7250521]
- Pellis SM, Field EF, Smith LK, Pellis VC. Multiple differences in the play fighting of male and female rats. Implications for the causes and functions of play. *Neurosci Biobehav Rev.* 1997; 21(1):105–120. [PubMed: 8994213]
- Pellis SM, Pellis VC. The prejuvenile onset of play fighting in laboratory rats (*Rattus norvegicus*). *Dev Psychobiol.* 1997; 31(3):193–205. [PubMed: 9386921]
- Philpot RM, Kirstein CL. The effects of repeated alcohol exposure on the neurochemistry of the periadolescent nucleus accumbens septi. *Neuroreport.* 1998; 9(7):1359–1363. [PubMed: 9631429]
- Philpot RM, Kirstein CL. Developmental differences in the accumbal dopaminergic response to repeated ethanol exposure. *Ann N Y Acad Sci.* 2004; 1021:422–426. [PubMed: 15251921]
- Piepponen TP, Kiianmaa K, Ahtee L. Effects of ethanol on the accumbal output of dopamine, GABA and glutamate in alcohol-tolerant and alcohol-nontolerant rats. *Pharmacol Biochem Behav.* 2002; 74(1):21–30. [PubMed: 12376149]
- Rodd ZA, Bell RL, McKinzie DL, Webster AA, Murphy JM, Lumeng L, Li TK, McBride WJ. Low-dose stimulatory effects of ethanol during adolescence in rat lines selectively bred for high alcohol intake. *Alcohol Clin Exp Res.* 2004; 28(4):535–543. [PubMed: 15100603]
- Sahr AE, Thielen RJ, Lumeng L, Li TK, McBride WJ. Long-lasting alterations of the mesolimbic dopamine system after periadolescent ethanol drinking by alcohol-preferring rats. *Alcohol Clin Exp Res.* 2004; 28(5):702–711. [PubMed: 15166644]
- Savage DD, Queen SA, Sanchez CF, Paxton LL, Mahoney JC, Goodlett CR, West JR. Prenatal ethanol exposure during the last third of gestation in the rat reduces hippocampal NMDA agonist binding site density in 45-day-old offspring. *Alcohol.* 1991; 9:37–41. [PubMed: 1346364]
- Slawecki CJ, Betancourt M. Effects of adolescent ethanol exposure on ethanol consumption in adult rats. *Alcohol.* 2002; 26(1):23–30. [PubMed: 11958943]
- Smith RF. Animal models of periadolescent substance abuse. *Neurotoxicol Teratol.* 2003; 25(3):291–301. [PubMed: 12757826]
- Smith GT, Goldman MS, Greenbaum PE, Christiansen BA. Expectancy for social facilitation from drinking: the divergent paths of high-expectancy and low-expectancy adolescents. *J Ab Psych.* 1995; 104(1):32–40.
- Smith AD, Weiss F. Ethanol exposure differentially alters central monoamine neurotransmission in alcohol-preferring versus -nonpreferring rats. *J Pharmacol Exp Ther.* 1999; 288(3):1223–1228. [PubMed: 10027862]
- Spear LP. The adolescent brain and age-related behavioral manifestations. *Neurosci Biobehav Rev.* 2000; 24:417–463. [PubMed: 10817843]
- Stansfield KH, Kirstein CL. Effects of novelty on behavior in the adolescent and adult rat. *Dev Psychobiol.* 2006; 48(10):10–15. [PubMed: 16381024]
- Stansfield KH, Kirstein CL. Chronic cocaine or ethanol exposure during adolescence alters novelty-related behaviors in adulthood. *Pharmacol Biochem Behav.* 2007; 86(4):637–642. [PubMed: 17395255]
- Strupp BJ, Levitsky DA. Social transmission of food preferences in adult hooded rats (*Rattus norvegicus*). *J Comp Psych.* 1984; 98(3):257–266.
- Substance Abuse and Mental Health Services Administration. NSDUH Series H-32. Office of Applied Studies; DHHS; Rockville, MD: 2007. Results from the 2006 National Survey on Drug Use and Health: National Findings.
- Szumilski KK, Diab ME, Friedman R, Henze LM, Lominac KD, Bowers MS. Accumbens neurochemical adaptations produced by binge-like alcohol consumption. *Psychopharmacology (Berl).* 2007; 190(4):415–431. [PubMed: 17225170]
- Thombs DL, Beck KH, Mahoney CA, Bromley MD, Bezon KM. Social context, sensation seeking, and teen-age alcohol abuse. *J School Health.* 1994; 64(2):73–79. [PubMed: 8028303]
- Tirelli E, Laviola G, Adriani W. Ontogenesis of behavioral sensitization and conditioned place preference induced by psychostimulants in laboratory rodents. *Neurosci Biobehav Rev.* 2003; 27:163–178. [PubMed: 12732232]

- Tolliver GA, Samson HH. The influence of early postweaning ethanol exposure on oral self-administration behavior in the rat. *Pharmacol Biochem Behav.* 1991; 38(3):575–580. [PubMed: 2068193]
- Truxell EM, Molina JC, Spear NE. Ethanol intake in the juvenile, adolescent, and adult rat: effects of age and prior exposure to ethanol. *Alcohol Clin Exp Res.* 2007; 31(5):755–765. [PubMed: 17386073]
- Vanderschuren LJMJ, Niesink RJM, Van Ree JM. The neurobiology of social play behavior in rats. *Neurosci Biobehav Rev.* 1997; 21(3):309–326. [PubMed: 9168267]
- Varlinskaya EI, Spear LP. Acute effects of ethanol on social behavior in adolescent and adult rats: Role of familiarity of the test situation. *Alcohol Clin Exp Res.* 2002; 26(10):1502–1511. [PubMed: 12394283]
- Varlinskaya EI, Spear LP. Ontogeny of acute tolerance to ethanol-induced social inhibition in Sprague-Dawley rats. *Alcohol Clin Exp Res.* 2006; 30(11):1833–1844. [PubMed: 17067347]
- Varlinskaya EI, Spear LP, Spear NE. Acute effects of ethanol on behavior of adolescent rats: Role of social context. *Alcohol Clin Exp Res.* 2001; 25(3):377–385. [PubMed: 11290848]
- Vetter CS, Doremus-Fitzwater TL, Spear LP. Time course of elevated ethanol intake in adolescent relative to adult rats under continuous, voluntary-access conditions. *Alcohol Clin Exp Res.* 2007; 31(7):1159–1168. [PubMed: 17511750]
- Weiss F, Lorang MT, Bloom FE, Koob GF. Oral alcohol self-administration stimulates dopamine release in the rat nucleus accumbens: genetic and motivational determinants. *J Pharmacol Exp Ther.* 1993; 267(1):250–258. [PubMed: 8229752]
- Weiss F, Parsons LH, Schulteis G, Hyttia P, Lorang MT, Bloom FE, Koob GF. Ethanol self-administration restores withdrawal-associated deficiencies in accumbal dopamine and 5-hydroxytryptamine release in dependent rats. *J Neurosci.* 1996; 16(10):3474–3485. [PubMed: 8627380]
- Yim HJ, Gonzales RA. Ethanol-induced increases in dopamine extracellular concentration in rat nucleus accumbens are accounted for by increased release and not uptake inhibition. *Alcohol.* 2000; 22(2):107–115. [PubMed: 11113625]
- Yoshimoto K, McBride WJ, Lumeng L, Li TK. Alcohol stimulates the release of dopamine and serotonin in the nucleus accumbens. *Alcohol.* 1992; 9:17–22. [PubMed: 1370758]

Figure 1.

The diagram is a schematic of the experimental protocol used in the social interaction experiment.

Preference for Chamber Paired with Social Interaction

Figure 2.

As depicted in the figure adolescent male alcohol naïve observer rats that socially interacted with demonstrators administered the highest dose of ethanol showed an increase in time spent in the chamber paired with social interaction with an alcohol-intoxicated demonstrator after conditioning relative to all other groups. Adolescent alcohol naïve female observers showed no change in time spent in the chamber paired with social interaction with an alcohol-intoxicated demonstrator, regardless of dose. Data presented are mean $\pm$ SEM difference in seconds spent in the paired chamber at test minus baseline presented as the change in time spent in the chamber paired with social interaction in the adolescent alcohol naïve observer's initially least preferred chamber. * indicates significant difference from all other sex- and dose-matched groups. The bars indicate the type of demonstrator the alcohol naïve adolescent observer socially interacted with (0.0 (clear bars)- social interaction with a sober peer; 0.5 (light grey) and 1.5 (dark grey)- social interaction with an alcohol-intoxicated peer). Female 0.0 $n = 13$; Female 0.5 $n = 14$; Female 1.5 $n = 13$; Male 0.0 $n = 8$; Male 0.5 $n = 11$; Male 1.5 $n = 10$.

Figure 3.

As depicted in the figure, animals that were pretreated with ethanol during adolescence and conditioned with ethanol in young adulthood spent more time in the ethanol-paired chamber relative to rats that were pretreated saline and conditioned with ethanol in young adulthood and also those that were pretreated with ethanol during adolescence and conditioned with saline in adulthood. Data presented are mean $\pm$ SEM of the change in time spent in the paired chamber in the initially least preferred chamber (test-baseline). The bars indicate conditioning in young adulthood with saline (clear bars) or ethanol (grey bars). Saline-Saline $n = 9$; Saline-Ethanol $n = 10$; Ethanol-Saline $n = 9$; Ethanol-Ethanol $n = 9$.

Figure 4.

In both panels, the x-axis denotes Age and the y-axis denotes extracellular DA levels (nM). Both approaches yielded similar age results [quantitative: $F(2, 12)=14.01$, $P < 0.05$; conventional: $F(3, 279)=17.171$, $P < 0.05$] with late adolescent animals (PND 45) having significantly higher basal DA levels than adults, but with the conventional method (Philpot and Kirstein, 2004), levels obtained in adolescents were higher than those obtained using the quantitative method (Badanich, Adler & Kirstein, 2006). Additionally, the quantitative approach revealed lower basal DA levels for early adolescent (PND 35) rats. For panel A, # = differs from PND 60; ## = differs from PND 45 and PND 60. For panel B, diamonds = differs from PND 45 and PND 60; stars = differs from all other ages. It should be noted the conventional method in panel B shows dialysate DA levels that were not corrected for probe recovery. Figures reproduced with permission from Philpot and Kirstein, 2004 and Badanich et al, 2006).

Figure 5.

The amount of DA in the microdialysis perfusate (DA_{in}) is on the x-axis while the difference between DA_{in} and recovered DA (DA_{out}) is on the y-axis (DA_{in} – DA_{out}). The broken horizontal line depicts the point of no net flux of DA between the brain and the microdialysis perfusate. The x-intercept denotes the extracellular DA concentration in the NAcc for ethanol-treated (solid line; n=4) and saline control (broken line n=4) rats. Basal DA was greater for rats pretreated with ethanol during adolescence (6.5 nM DA) than for saline controls (3.6 nM DA); [t(6)= 2.458, P < 0.05]. The slope of the regression line denotes the extraction fraction (E_d), an indirect measure of DA reuptake, for ethanol-treated (87%) and saline control (68%) rats. Each data point denotes mean and SEM. This figure has been reproduced with permission from Badanich et al., 2007.