Anxiety disorders are commonly reported in childhood and adolescence with prevalence rates between 5.3% and 17% (Cartwright-Hatton, McNicol, & Doubleday, 2006). For a significant number of these children and adolescents, these anxiety problems can persist into adulthood (Pine, Cohen, Gurley, Brook, & Ma, 1998). The principles of association described by learning theory have long been used to explain how anxiety problems develop (e.g., Watson & Rayner, 1920). However, most studies investigating the nature of fear learning difficulties in anxious and nonanxious individuals have focused on adults when presumably much of the learning related to the anxiety response has already taken place.

Fear learning in experimental settings is commonly assessed using Pavlovian conditioning procedures, the process in which a neutral stimulus (CS+) is repeatedly paired with a frightening stimulus (US), such that the neutral stimulus acquires a fear-provoking value. Conditioning is often found to be more effective with repeated pairings of the neutral stimulus (or situation) with the aversive event (or outcome). However, one-trial learning in rats and humans (e.g., Garcia, McGowan, & Green, 1972; Öhman, Eriksson, & Olofsson, 1975) shows that an association between a neutral stimulus can also be easily acquired with a single traumatic event. As not everyone exposed to such an experience develops an anxiety disorder, contemporary learning theories of anxiety assume a diathesis stress model in which conditioned experiences only result in anxiety responses in individuals who are particularly vulnerable (Mineka & Zinbarg, 2006), possibly because of an inherited predisposition (Hettema, Annas, Neale, Kendler, & Fredrikson, 2003) or acquired through social learning from anxious parents (Field & Lester, 2010). This inherited/acquired vulnerability may manifest through impairments in learning: Research in human adults has shown that anxious individuals (1) respond with higher fear levels to a newly acquired CS+ compared with nonanxious adults (Lissek et al., 2005), (2) exhibit heightened fear reactions in response to stimuli not paired with the US (CS−) seeming to overgeneralize fear from CS+ to CS− (Lissek et al., 2005), and (3) more tentatively,
respond with greater fear to the context in which fear associations are formed (Baas, 2012; Grillon, 2002).

Differences in acquisition of fear are accompanied by differences in the loss or extinction of fear. Here, the neutral stimulus is presented without the frightening stimulus over several trials to allow for either a reduction in excitatory association or a new association with safety to be formed; in either case, the CS+ loses its fear-provoking value, and anxiety is usually reduced. Thus, fear learning can also be applied to understand anxiety reduction, and exposure therapy that relies on extinction principles is an integral part of most anxiety treatments (Anderson & Insel, 2006; Delgado, Olsson, & Phelps, 2006). However, again, not all individuals who have experienced the same traumatic events show a reduction in fear across time – and indeed, it may be that those with clinical anxiety seek help because extinction has not occurred naturally. This is consistent with empirical data showing that anxious patients have greater difficulties extinguishing fear (e.g., Michael, Blechert, Vriends, Margraf, & Wilhelm, 2007).

Comparably less is known about how disruptions in fear learning and extinction can explain persistent fears and worries in childhood and adolescence. In this chapter, we focus on two key questions looking at evidence from human and animal models: (1) Individual differences, that is, are some children and adolescents more prone to anxiety than others because of difficulties in fear learning and extinction, and what might the neural basis be using studies of functional magnetic resonance imaging (fMRI)? (2) Qualitative developmental change, that is, does the nature of fear learning change across age through experience-dependent maturation of the PFC and amygdala using lesion studies in animals and fMRI studies in humans – and can this explain why anxiety typically onsets in adolescence?

**Individual Differences in Human Fear Learning**

Fear-conditioning and extinction paradigms can be divided into two types: (1) simple fear-conditioning paradigms, where a neutral stimulus is paired with an unconditioned stimulus (UCS), thereby becoming a conditioned threat stimulus (CS+); and (2) simple differential fear-conditioning paradigms, where two neutral stimuli are presented. One stimulus is paired with the UCS (CS+), and a second stimulus is never paired with the UCS (CS–). Thereby, the CS− acquires a conditioned safety value. In the first of these paradigms, CRs to the CS+ alone are measured during (or after) conditioning and during (or after) extinction. In the second paradigm, CRs to both the CS+ and CS– can be measured across phases in addition to their difference.

Studies of fear conditioning in anxious and nonanxious youth

Clinically anxious adults have been found to show greater CRs to the CS+ compared with nonanxious adults in simple conditioning. However, results from studies using differential conditioning paradigms have been less consistent. Often, greater CRs to the CS+ and CS– in anxious than nonanxious individuals have been found with no significant group differences in the differential CR to the CS+ relative to the
CS− responses (Lissek et al., 2005). Only five studies (Table 18.1) have explored the relationship between anxiety and fear learning in youth, with the majority reporting group differences – however these have varied over where the group differences lie. Perhaps the most consistent finding is that indices of conditioned fear responses, for example, skin conductance responses (SCR) and verbal fear ratings, are higher in clinically anxious children and adolescents to the CS+ (Craske et al., 2008; Waters, Henry, & Neumann, 2009), similar to findings from the adult literature (Lissek et al., 2005).

Also, similar to adult findings, anxious children and adolescents appear more afraid of the CS− too (Craske et al., 2008; Lau et al., 2008; Waters et al., 2009; but see Liberman, Lipp, Spence, & March, 2006). This means that in general, there are no group differences found in differential conditioning (the difference between responses to the CS+ and CS−). Thus, these studies suggest that anxious, like nonanxious, youth can differentiate fear to the CS+ and the CS− (though see Liberman et al., 2006) but that anxious youth manifest enhanced fear to the CS+ that generalizes to the CS−. This could imply sensitization (enhanced fear to all experimental stimuli and the wider context) but could also occur because of an inability to discriminate between stimuli that are perceptually similar. These questions have been investigated in anxious adults (Haddad, Pritchett, Lissek, & Lau, 2012; Lissek et al., 2005) but not yet in anxious children and adolescents. Of note, studies of children and adolescents do not typically employ electric shock as the UCS – instead relying on more mildly aversive stimuli, such as loud noises. A possible reason for the more mixed findings in the child and adolescent literature is that these UCSs are not sufficient in producing conditioned fear – this possibility is explored in more detail below.

Studies of fear extinction in anxious and nonanxious youth

In adults, meta-analyses have found that overall, compared with nonanxious controls, anxious individuals show stronger fear responses to the CS+ during extinction in simple conditioning paradigms. However, no differences between anxious and nonanxious participants emerge when comparing the magnitude of the difference in fear to the CS+ versus CS− in differential conditioning paradigms (similar to at the end of acquisition; Lissek et al., 2005). Studies investigating extinction in highly anxious children and adolescents have again yielded mixed findings. One study found a higher fear response in anxious children and adolescents to the CS+ (Waters et al., 2009), whereas another study found the opposite with a higher fear response in nonanxious children and adolescents (Craske et al., 2008).

Findings on differential conditioning are similarly inconclusive. While there is evidence that during extinction, there are within-group differences to the stimuli in all participants, that is, both anxious and nonanxious children and adolescents are more afraid of the CS+ than the CS− (Lau et al., 2008), there is also evidence that only anxious children and adolescents display differential fear responses (Liberman et al., 2006; Waters et al., 2009). Still other studies have reported an absence of differential SCR during extinction in both anxious and nonanxious children and adolescents – but that anxious individuals were generally more afraid of both the CS+ and CS− compared with nonanxious children and adolescents (Craske et al., 2008; Liberman et al., 2006; Waters et al., 2009).
Table 18.1  Fear learning in humans.

<table>
<thead>
<tr>
<th>Authors</th>
<th>Age</th>
<th>Anxiety measure</th>
<th>Outcome</th>
<th>Acquisition</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Individual differences in anxiety</td>
<td></td>
<td></td>
<td>CS+</td>
<td>CS–</td>
<td>US</td>
</tr>
<tr>
<td>Craske et al. (2008)</td>
<td>7–12.9</td>
<td>Clinical severity rating of 4 or higher for separation anxiety disorder, panic disorder, generalized anxiety disorder, or social anxiety disorder or specific phobia with a CSR of 4 or greater if accompanied by another anxiety disorder diagnosis with a CSR of 3</td>
<td>SCR, verbal (arousal ratings)</td>
<td>Trapezoid Triangle Aversive tone N/A 16</td>
<td>CS+ was rated as significantly less pleasant by the anxiety than control group; no difference in their ratings of the CS–</td>
</tr>
<tr>
<td>Liberman et al. (2006)</td>
<td>7–14</td>
<td>Meeting clinical diagnostic criteria for one or more anxiety disorders</td>
<td>Verbal, SCR, FPS Picture picture Loud tone N/A 12</td>
<td>Differential conditioning only in nonanxious group (fear)</td>
<td>Anxiety group did not differ from nonanxiety group; No differential conditioning</td>
</tr>
<tr>
<td>Authors</td>
<td>Age</td>
<td>Anxiety measure</td>
<td>Outcome</td>
<td>Acquisition Results</td>
<td></td>
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<tr>
<td>Waters et al.</td>
<td>8–12</td>
<td>Clinical severity rating of 4 or higher social phobia, generalized anxiety disorder, or specific phobia</td>
<td>Verbal SCR</td>
<td>CS+ Trapezoid/ triangle</td>
<td>CS– Trapezoid/ triangle</td>
</tr>
<tr>
<td>Lau et al.</td>
<td>13.64 (2.37)</td>
<td>Meeting clinical diagnostic criteria of the DSM-IV for an anxiety disorder</td>
<td>Verbal Face1</td>
<td>Face1 (scared) + scream</td>
<td>Face2</td>
</tr>
<tr>
<td>Study</td>
<td>Age Range</td>
<td>Stimuli</td>
<td>Condition</td>
<td>SCR</td>
<td>Tone 1</td>
</tr>
<tr>
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<tr>
<td>Gao et al. (2010)</td>
<td>3–8</td>
<td>SCR</td>
<td>Tone 1</td>
<td>Tone 2</td>
<td>Loud Tone</td>
</tr>
<tr>
<td>Pattwell et al. (2012)</td>
<td>5–11</td>
<td>SCR</td>
<td>Square 1</td>
<td>Square 2</td>
<td>Loud Tone</td>
</tr>
<tr>
<td>Glenn, Lieberman, and Hajcak (2012)</td>
<td>8–10</td>
<td>Verbal, FPS</td>
<td>Face 1</td>
<td>Face 2</td>
<td>Scream + Face 1 (scared)</td>
</tr>
</tbody>
</table>

(Continued)
<table>
<thead>
<tr>
<th>Authors</th>
<th>Age</th>
<th>Anxiety measure</th>
<th>Outcome</th>
<th>CS+</th>
<th>CS–</th>
<th>US</th>
<th>%</th>
<th>No.</th>
<th>Acquisation</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lau et al. (2011)</td>
<td>10–17</td>
<td>18–50</td>
<td>Verbal (E1&amp;2), SCR E1, BOLD(E2)</td>
<td>Face1</td>
<td>Face2</td>
<td>Scream + Face1 (scared)</td>
<td>20/60</td>
<td>80/50</td>
<td></td>
<td>Experiment 1: GSR: differential conditioning in both groups; overall, higher responses in adolescents Verbal: differential conditioning, no age differences Experiment 2: Verbal: differential conditioning in both group, stronger differential conditioning in adults BOLD: differential BOLD response in right hippocampus in adolescents and adults, differential BOLD response in right amygdala and left hippocampus only in adolescents, greater activity in DLPFC correlated with higher fear ratings to the CS– in adults, but lower activity predicted more fear to the CS– in adolescents</td>
</tr>
</tbody>
</table>
Clearly, the evidence of differences in either fear learning or extinction between anxious and nonanxious youth is mixed. One reason for the inconsistencies is that there is a paucity of fear-conditioning studies in children and adolescents – and therefore the inconsistent results in this small number of studies could be attributed to methodological differences between the studies. Studies use quite different fear indices, and there is a lack in standardization of the conditioning protocol. Additionally, studying fear processes in youth requires balancing practical and ethical considerations. Electrical shocks, the most powerful UCS in adults, are not appropriate for adolescents. Less noxious UCSs however, such as loud auditory stimuli or shocking or unpleasant photographs, while useful in working with children, provoke minimal fear in the adolescent age range (Lau et al., 2008). To tackle this problem, a novel paradigm has recently been introduced that uses a piercing female scream as the aversive UCS. The “screaming lady paradigm” has been successfully used in both healthy and clinical populations (Lau et al., 2008, 2011).

A further drawback is that research on fear learning during development to date has used discrete cue conditioning, a paradigm that is best suited for explaining transient fear states in both anxious and nonanxious individuals. Context conditioning or conditioning to diffuse nonspecific “background” cues has been used to explain situations of more generalized and sustained fear responses, in other words, anxiety. Contextual fear may be related to the background context during which an aversive stimulus was experienced or acts as a moderator of the effects of the exogenous threat cue itself. Previous work with adults suggests that this contextual fear is greater under conditions when the CS/UCS association is less predictable, that is, when the UCS does not necessarily follow the CS (Grillon, Baas, Cornwell, & Johnson, 2006). This draws on earlier animal work that demonstrated that the CS is unlikely to elicit a CR during the testing stage if, during training, the UCS had a higher probability in the absence than in the presence of the CS (Rescorla, 1968) – probably because the context attained a higher predictive value than the CS (Goddard & Jenkins, 1987). Recent work with anxious adults has shown that this contextual fear response under unpredictable circumstances is even more enhanced in high-anxious individuals (Baas, 2012). In a recent study, Kadosh and colleagues (2015) investigated developmental differences in threat learning in different context conditions in a sample of high- and low-anxious adolescents (aged 13–18). They showed that high-anxious adolescents failed to establish a discriminate response between threat and safety cues, by overgeneralizing fear responses from the CS+ to the contexts in which they appeared. This finding led the authors to suggest that high trait anxiety early in development may be associated with an inability to discriminate cues and contexts, and a misunderstanding of safety or ambiguous signals.

Finally, specific fear learning deficits may explain anxiety during some stages of development but not during others. More particularly, if fear conditioning and extinction rely on certain brain regions that are undergoing structural and functional maturation from late childhood and across adolescence to early adulthood, perhaps the case with which conditioned fear arises and abates, and the extent to which it explains individual differences in anxiety changes across these developmental phases. The next section will consider age-associated changes in the sensitivity to different
fear-learning indices including during acquisition and extinction – and retention of these learned associations. As most of the work has been conducted in rodents, these will be reviewed first.

### Developmental Changes in Fear Learning

#### Nonhuman animal work

There has been a longstanding tendency to use animal subjects for the study of fear learning, starting with Pavlov (1927). Animal subjects offer unique options in methodology, and not surprisingly, rodents have become the most commonly used subjects in recent years. Rodent studies have several advantages. For example, novel drugs can be administered systematically (Milad & Quirk, 2012), brain areas can be lesioned, and brains can be dissected postmortem to gain a better understanding of the underlying neuronal circuitry. The neural circuits involved in adult fear acquisition and extinction have been found to be comparable in rodents and humans (Graham & Milad, 2011). Even though prefrontal areas in humans are more developed than in rats (Milad, Rauch, Pitman, & Quirk, 2006), the prelimbic (PL) and infralimbic (IL) regions in rodents have been established as homologs to the human medial PFC (Milad & Quirk, 2012). This cross-species validity allows one to translate findings from rodent studies to understand human processes. Crucially, studies of how fear learning develops with age can also benefit from this translational work, given that rats and humans undergo similar developmental stages. Postnatal day (P) 16 in rats is comparable with human infancy. At P24, a rat is a juvenile (or preadolescent), while P28 and P35 correspond approximately to early and late adolescence respectively, and P70 corresponds to adulthood.

#### Studies of fear conditioning

Previous research suggests that the capacity to learn fear-relevant associations develops gradually across infancy, first appearing at the age of P10 (Figure 18.1). In two studies, rats at various stages in infancy were exposed to an odor that was paired with a shock (Sullivan Landers, Yeaman, & Wilson, 2000; Thompson, Sullivan, & Wilson, 2008) and subsequently tested on a two-odor choice test. In this test, rats were placed in a Y-maze and had to choose to walk toward either the conditioned or another familiar odor. At P8, rats displayed a preference for the conditioned odor, indicating that acquisition was probably unsuccessful and that rats of this age had not learned to fear the odor despite being paired with a UCS. In contrast, from P10, rats were able to learn to avoid an aversive stimulus; and moreover, by P12, the two-odor choice test revealed that this conditioned avoidant response lasted at least 4 hr and even 24 hr after acquisition.

At P16, as few as two pairings were sufficient for a rat to learn to fear (indexed by freezing behavior) a CS when tested immediately after acquisition (Kim, Li, Hamlin, McNally, & Richardson, 2012); and at the beginning of extinction, 7 or 8 days after acquisition (Kim & Richardson, 2010; Yap, Stapinski, & Richardson, 2005). However,
even though fear learning appears to be present at P16, crucial differences between P16 and older rats have been observed. For instance, several studies administered more pairings to P16 rats than the older rats to obtain comparable levels of fear (e.g., Kim, Hamlin, & Richardson, 2009) – and P16 rats also show greater spontaneous loss of responding (perhaps related to forgetting; see next section).

By P28 (early adolescence), fear learning can be reliably generated, although more subtle changes have been documented. Hefner and Holmes (2007) found enhanced fear acquisition in P28 mice compared with adult mice, but by P35 these age-associated differences disappeared (Kim et al., 2011; McCallum, Kim, & Richardson, 2010). To investigate this effect further, Den and Richardson (2013) compared delayed and trace conditioning in P23, P28, and P35 rats. During delayed fear conditioning, the CS+ and UCS overlap in time, while in trace fear conditioning, CS+ offset and UCS onset are separated by several seconds, a procedure that makes it more difficult to learn the association between CS+ and US. While neither P23 nor adult rats were able to acquire fear learning when the CS+ and UCS were separated by 20 or 40 s, P35 rats showed successful acquisition under both conditions, with freezing rates comparable with delay fear conditioning. Taken together, these data suggest that between P28 and P35, rats may be more sensitive in detecting the relationships between the neutral and the aversive stimuli.

An important caveat to note when interpreting these results is that appropriate measures of conditioned fear may also depend on age. For example, fear-potentiated startle (FPS) develops later than freezing and avoidance (Richardson, Fan, & Parnas, 2003; Richardson, Paxinos, & Lee, 2000; Richardson, Tronson, Bailey, & Parnas, 2002). By P16 and P20, rats show successful fear learning by avoiding a CS paired with shock but equal levels of FPS to the unpaired CS as to the paired CS. At P23 and P75, learned fear is evident when indicated by either avoidance or FPS (Richardson et al., 2000). These data underscore the need for developmentally appropriate measurement tools to investigate age-associated changes in fear learning.

The behavioral changes in fear learning across infancy, adolescence, and adulthood are accompanied by changes in neural activity of relevant brain regions. The amygdala has been consistently implicated in fear conditioning (Milad & Quirk, 2012). In early development, when rats do not yet show fear learning, they also do not display neural activity in the amygdala during fear acquisition – coinciding with decreased levels of synaptic plasticity in the basolateral amygdala (BLA) at P8 (Thompson et al., 2008).
However, from P10 onwards, increased neural activity in the amygdala emerges in response to acquisition, and synaptic plasticity is also observed in the BLA. Interestingly, if synaptic plasticity in the amygdala is disrupted by blocking gamma-aminobutyric acid (GABA) receptors in P12 rats, fear conditioning is also disrupted (Sullivan et al., 2000; Thompson et al., 2008). The development of synaptic plasticity may be related to N-methyl-d-aspartate (NMDA) receptors, which play an important role in controlling synaptic plasticity in adulthood. Injecting P16 and P23 rats with MK-801, an NMDA antagonist, during acquisition similarly impairs fear acquisition (Langton, Kim, Nicholas, & Richardson, 2007).

The medial PFC (mPFC), particularly the infralimbic cortex (IL) and the prelimbic cortex (PL), also play important roles in the modulation of amygdala activity during rodent fear learning (Quirk & Beer, 2006). The PL in particular has been found to be important for fear expression, whereas the IL is more involved in fear inhibition (Sotres-Bayon & Quirk, 2010). At P23, fear acquisition involves an enhancement of synaptic transmission at the PL glutamatergic synapses, but by P29 this synaptic transmission did not change in response to acquisition (Pattwell et al., 2012).

Studies of spontaneous forgetting and reactivation

As mentioned above, although there is evidence that P16 rats show fear learning, there may be memory differences compared with P24 rats, such that P16 rats display spontaneous loss of responding or forgetting after acquisition. P16 rats show substantially lower levels of freezing in response to the CS if tested 48 h after acquisition compared with an immediate test (Kim et al., 2012). This spontaneous decrease in the CR does not characterize P24 rats. These findings have been supported by another study that also found that P23, but not P16, rats displayed heightened fear levels to the CS+ 2 days after acquisition. Thus, even though rats can acquire a CS+–US relationship at P16, they seem less efficient in retaining learned fear (Kim & Richardson, 2007a; Kim et al., 2012) unless they receive more pairings of the CS+ and UCS (e.g., six acquisition trials rather than just two). There is evidence, however, that even when P16–P17 rats show signs of spontaneous forgetting, the memory does not seem to be completely lost over time. That is, using a process called reactivation or reinstatement (Bouton, 2002), where reminder shock is administered 1 day before testing, learned fear can be successfully elicited 3–7 days after acquisition (Kim & Richardson, 2007a; Li, Kim, & Richardson, 2012b).

These age differences in the expression of the fear memory appear to be independent of amygdala functioning. For example, Kim et al. (2012) found that although only the older (P23) rats showed higher levels of freezing toward the CS+ postacquisition, there was elevated phosphorylated mitogen-activated protein kinase (pMAPK)-immunoreactive neuronal activation in the amygdala in both P16 and P23 rats. In P16 rats who showed improved acquisition memory after six CS–US pairings, the pMAPK count was equally high in the group that received six, two, and no pairings. In contrast, differences in the expression of acquired fear may be reliant on the prelimbic (PL) region of the vmPFC. Following PL inactivation (which was achieved by injecting muscimol, a GABAergic agonist), P23 rats behaved like P16 rats with lower levels of freezing (Li et al., 2012a). Together, these findings
lend support to the notion that the PL is not crucial for the expression of learned fear at P16 but becomes critical at P23.

Extinction and extinction retention

In contrast to the acquisition of fear, extinction (i.e., when the CS is no longer paired with the UCS) appears to vary less with age, with similar declines in fear-expression rates (as measured by freezing) being reported in P16 rats as P24 rats (e.g., Langton et al., 2007; McCallum et al., 2010). We note that this does depend on the number of extinction trials presented, with most studies reporting successful extinction across 30 trials but not five (Pattwell et al., 2012).

However, when it comes to maintaining acquired knowledge, there are age-associated changes. Successful extinction retention 24 hr after extinction has been found in rats at P70 (adulthood; McCallum et al., 2010) but also earlier on in development, at P16/17 and P23/24 (e.g., Langton et al., 2007). These effects are strikingly persistent with low levels of freezing, hence successful extinction retention, continuing to characterize P16 rats even after 6–7 days postextinction learning (Kim & Richardson, 2010; Yap & Richardson, 2007). Interestingly, the retention of extinction was impaired in adolescent rats (Kim, Li, & Richardson, 2011; McCallum et al., 2010) and mice (Pattwell et al., 2012) compared with preadolescent and adult animals—and only emerged under two conditions: (1) when the extinction experience was doubled (Kim et al., 2011; McCallum et al., 2010) or (2) when d-cycloserine (DCS), an NMDA partial agonist, was administered immediately after extinction (McCallum et al., 2010). DCS has been found to facilitate extinction in adult rats (Ledgerwood, Richardson, & Cranney, 2003).

Thus, while adolescent rats and mice show normal within-session extinction (Hefner & Holmes, 2007; Kim et al., 2011), extinction retention appears to be attenuated in this age range (Kim et al., 2011; McCallum et al., 2010). As with fear conditioning, these behavioral changes related to extinction retention during development occur in tandem with changes in the engagement of neural circuits, possibly because certain brain regions reach maturity at different stages. Most studies have noted similar engagement of the amygdala during extinction learning (Kim et al., 2009). However, age-associated changes during extinction learning have been reported in the vmPFC, particularly in the infralimbic (IL) region (Kim et al., 2009), which, in adult rats, has been found to be involved in mediating extinction acquisition and retention by inhibiting central amygdala responses to suppress fear expression (Sotres-Bayon & Quirk, 2010). In a series of studies conducted by Kim et al. (2009), the pMAPK count in the IL, and to some extent in the PL, was found to be elevated in P24 rats in response to extinction learning but not in P17 rats. pMAPK is an enzyme that is part of the intracellular signaling pathway and is important for activity-dependent modulation of synaptic strength. Furthermore, inactivating the mPFC before extinction severely impaired extinction retention in P24 rats but had no effect in P17 rats. Together, these data imply that only at P24 do rats rely on mPFC for extinction retention.

Other changes also occur in the role of the IL and the PL during the retention of extinction during the adolescent years. For example, Pattwell et al. (2012) found that,
in line with Kim et al. (2009), IL activity increased, and PL activity decreased in P23 and adult mice but not in P29 1 day after extinction (compared with control groups who did not receive extinction training). In P23 and adult mice, these changes in activity were also accompanied by an enhancement of glutamatergic synaptic transmission in the IL L5 pyramidal neurons. As with behavioral findings, when the adolescent rats received 60 trials of extinction instead of 30, not only was extinction retention improved but pMAPK counts in the IL and PL were higher than in rats that received no extinction or 30 extinction trials only. Thus, adolescent rodents are able to engage the IL and PL during extinction retention if extinction is increased. Together, these data imply less efficient neural networks in adolescent rodents (Kim et al., 2011).

Return of fear

Originally, it was assumed that successful extinction leads to the erasure (or unlearning) of the fear memory (Rescorla & Wagner, 1972). However, since then, a vast number of studies have shown that under the appropriate circumstances fear returns (e.g., spontaneous recovery; Quirk, 2002). The most common paradigms to study the return of fear are renewal, reinstatement, and spontaneous recovery (Bouton, 2002). Renewal refers to the process in which fear returns in a context different from extinction. This effect is particularly strong when the subject is returned to the context in which acquisition took place. Reinstatement is when a return of the fear response appears after extinction when subjects are presented with the US alone (reinstatement). Spontaneous recovery refers to the finding that the mere passage of time after extinction leads to reemergence of conditioned fear. Fear also commonly returns after extinction when the CS+ is presented in a context other than the extinction context – classically the acquisition context. Collectively, these phenomena of the return of fear suggest that extinction leads to new learning as opposed to memory erasure.

Several studies now show that renewal does not occur in P16 rats. For instance, one study systematically controlled the context in which fear learning took place. As a result, one group of rats received acquisition, extinction, and testing in the same environment (AAA). Others received acquisition in one context, and extinction and testing in another (ABB). In both cases, extinction and testing context were identical. Rats in the renewal condition either were placed into context A during acquisition, then placed into context B during extinction and returned to context A for testing (ABA), or received acquisition and extinction in the same context but placed in another context for testing (AAB). These last two conditions are considered examples of renewal, as the extinction and testing context were different. While P16 and P23 rats show extinction retention to a similar extent in the AAA and ABB condition, only P23 rats show renewal in the ABA condition. This lack of return of fear in P16 rats could indicate that at this age, extinction may look more like the erasure of the acquisition memory.

Similar to the findings on renewal, reinstatement does not appear to be present in rats younger than P23 (Callaghan & Richardson, 2011; Kim & Richardson, 2007b). P23 rats showed reinstatement in response to a US reminder in the form
of a postextinction shock. Their freezing levels were elevated compared with rats that did not receive a reminder. P16 rats, on the other hand, showed equally low levels of freezing in the reminder and no-reminder group, and hence displayed no reinstatement. Of note is the fact that P23 rats did not show return of fear when the reminder was presented in a context different from the context in which extinction and testing took place. Thus, reinstatement was modulated by the context in preadolescent rats.

Results for spontaneous recovery are more mixed. One study observed increased freezing levels in response to the CS+ 7 days after extinction in P23 mice, while P16 mice displayed substantially lower levels of freezing. This indicates successful extinction but simultaneously can be interpreted as the absence of spontaneous recovery (Gogolla, Caroni, Lüthi, & Herry, 2009). In contrast, Pattwell et al. (2012) observed only slight increases in freezing 24 h after extinction in P29 and adult mice but not at P23. However, the experimental procedures involved in these studies were very different, especially over the delay between extinction and testing.

Taken together these findings show that P16 rats fail to exhibit renewal, reinstatement, and spontaneous recovery suggesting that at this developmental juncture, extinction may well erase fear memories more permanently. It appears that, whereas new learning takes place in P23 and adult rats, unlearning takes place in P16 rats. The fear memory is permanently erased. Alternatively, contextual manipulations in these experiments could be less effective for younger rats.

With regards to the underlying brain networks, it has been shown that the amygdala is a crucial brain structure in both acquisition and extinction from P10. Interestingly, there is an increase in perineuronal nets in the BLA between P16 and P21, which has been interpreted as evidence that perineuronal nets protect the fear memory from being overwritten by extinction. Also, consistent with this interpretation: When perineuronal nets were destroyed in adult mice, these mice resembled P16 mice with a failure to exhibit renewal or spontaneous recovery (Gogolla et al., 2009).

**Humans**

Studies of fear conditioning

Table 18.1 also displays studies comparing fear learning across development. As with rodents, differential conditioning has been found in young children as early as 3 years (Gao et al., 2010). Unlike rodent studies, the evidence for age-associated differences in the learning of fear associations is less convincing. Only one study has reported such differences: Comparing 8- to 10-year olds and 11- with 13-year olds, this study reported greater differences between CS+ and CS– in the older age group using FPS (Glenn, Klein, et al., 2012). In other studies, one study with an age range of 5–28 found that age had no effect on the SCR in response to either CS+ or CS– (Pattwell et al., 2012), and in another, differential SCR to the CS+ and CS– did not vary in adolescence (10 to –17 years) relative to adults (18–50 years), although overall greater fear responses emerged in the adolescent group.
Studies of fear extinction

In terms of extinction, again mirroring rodent studies, preadolescent children appear capable of reducing their fear to a previously fearfult stimulus (Neumann, Waters, Westbury, & Henry, 2008). Moreover, this acquired fear reduction appeared no different to that found in adults (Pattwell et al., 2012): Thus, both groups displayed a strong decrease in SCR from the first to the last extinction trial. Interestingly, this study, which also included an adolescent group, showed that within-session extinction was clearly attenuated in adolescence. If replicated, these findings show a good parallel to rodent studies: notably that extinction in humans (and retention of extinction in rodents) is more problematic in the adolescent years compared with childhood or adulthood – a finding that carries implications for the understanding of why there may be an onset of persistent anxiety in adolescence.

Section summary

The nature of fear learning changes dramatically throughout life (see Table 18.2 and Figure 18.1), possibly driven by a combination of maturational and experience-dependent processes. Infant rats show associative learning during both the acquisition and extinction of fear, but it is clear from retention studies, notably studies of spontaneous forgetting and the return of fear after extinction, that as juveniles, these fear memories are not stable. The poorer capacity to retain learned fear associations may arise from developmental differences in amygdala functioning, which have been attributed to developmental immaturity of this region. Another shift in fear learning occurs in the transition across adolescence. During this period, fear is reliably acquired, but there appears to be a greater sensitivity for acquiring fear-relevant associations. In addition, while there are no age-associated differences in extinction learning, preliminary data are suggestive of adolescent-specific declines in the retention of extinguished fears. Thus, in contrast to juvenile and adult rodents, adolescent rodents require more extinction trials (or pharmacological agents) – a finding that may be mediated by immature medial PFC engagement.

How do these rodent findings map onto age-associated differences in human fear learning? The paucity of studies comparing different age groups in fear acquisition and extinction makes drawing parallels difficult – but there is tentative evidence that adolescents may show greater acquired fear to threat cues (relative to safety cues), while extinction learning is more difficult to acquire. If these findings are replicated, this may provide a plausible reason why adolescence is a period associated with the onset of more persistent forms of anxiety. It is also interesting to note that as structures such as the medial PFC are involved in processes such as extinction learning (Phelps, Delgado, Nearing, LeDoux, 2004) – and that such structures are still maturing in adolescence (relative to subcortical structures such as the amygdala; Casey et al., 2008), this may explain the observed differences in fear learning particularly extinction. Clearly, these suggestions are based on a limited number of behavioral studies in humans, but as many rodent studies find more convincing developmental differences in general and adolescent-associated differences in particular in studies that examine the retention of these fear memories, this should be an avenue for future studies to explore.
Table 18.2  Fear learning in childhood and adolescence, a cross-species comparison.

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*Only tested for avoidance as outcome.
Conclusions

This chapter has reviewed the nature of fear learning in children and adolescents, examining both how fear learning difficulties may characterize anxious and nonanxious youth, and the emergence of the associative processes related to fear-learning capacity and the underlying neural substrates across age. Although the limited number of studies makes drawing strong conclusions premature, it is clear that fear learning may play some role in explaining why some children and young people develop anxiety problems. However, fear learning may also explain why many persistent anxiety disorders first emerge in adolescence. The vital clue involves examining the nature of fear learning in childhood, adolescence, and adulthood – and there is now an emerging corpus of data (mostly from rodent studies) that suggest enhanced sensitivity to acquiring fear associations in adolescence and difficulties acquiring/retaining these associations after extinction.

These findings have clear implications for understanding the developmental time course not only of anxiety, but also of its treatment. In humans, extinction is assumed to be the underlying mechanism of exposure therapy (Rothbaum & Davis, 2003), and successful extinction is a potential predictor of treatment success. Adults that show better retention of extinction also improve more in social anxiety symptoms following exposure therapy (Berry, Rosenfield, & Smits, 2009). The presented research suggests that exposure therapy might be effective for childhood anxiety but less successful for adolescent anxiety. These findings point toward ways to enhance exposure therapy, for example by extending the number of sessions or introducing pharmacological interventions, which might facilitate exposure treatment in adolescents, such as the NMDA agonists DCS (McCallum et al., 2010). In human adults, DCS has been found to be a promising way to enhance exposure therapy (Byrne, Farrell, & Rapee, 2011). Alternatively, anxious adolescents may benefit more from other forms of psychological treatments such as cognitive therapy or more recently developed cognitive bias modification training programs (Lau, 2013).

References


