Is empathy involved in our emotional response to music? The role of the PRL gene, empathy,

and arousal in response to happy and sad music

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Abstract

Recent studies have shown that empathy is a potentially important factor in understanding the emotional impact of music. The aim of this study was to explore associations between empathy and felt/perceived emotions while listening to music. The assessment of empathy was undertaken using the Interpersonal Reactivity Index (IRI). As earlier work has suggested that prolactin could be an important hormone in enhancing empathy while listening to sad music, we investigated whether two genetic polymorphisms located on the PRL gene (coding for prolactin) could explain individual differences in reactions to listening to music. N = 160participants were recruited for this study. All participants provided buccal swabs for genetic analysis. All participants listened to 10 sad and 10 happy musical excerpts, and after each song they reported the emotion they felt or perceived. Several significant associations appeared between empathy and the felt/perceived emotions while listening to music. With respect to the genetic markers, an effect of one prolactin polymorphism (rs1205960) was shown. TT/TC carriers reported significantly lower arousal levels compared to the CC carriers after having heard the happy and sad music. The results from this study showed that i) empathy is involved in the process of perceiving and feeling emotions while listening to music and ii) prolactin might play a role in eliciting different emotional reactions, based on arousal level, while listening to happy or sad music. Finally, we report non-significant findings in relation to three SNPs from the oxytocin receptor gene (OXTR), which are presented in the supplementary material.

Keywords: empathy, music, emotions, PRL_rs1205960, PRL_gene

Introduction

Empathy, which can be defined as the ability to experience the emotional state of others (affective empathy) and to put oneself in the shoes of another person (cognitive empathy), is essential for successful and fulfilling social interactions since it determines the appropriateness of an individual's reactions to the emotional states displayed by others (Melchers, Montag, Markett & Reuter, 2013). Deficits in empathy are often considered a hallmark of a range of psychological disorders (Greenberg, Rentfrow & Baron-Cohen, 2015; Melchers, Li, Chen, Zhang & Montag, 2015). In particular, autism spectrum disorder (ASD) is often associated with deficits in empathy, mainly relating to the ability to infer emotions from facial expressions (Baron-Cohen et al., 2015).

Emotions are not, however, exclusively communicated via our facial expressions. The human voice and music (which can include vocal elements) can be used to convey information about emotional states (e.g. Hunter, Schellenberg & Griffith, 2011, Schellenberg & Scheve, 2012, Rolison & Edworthy, 2013). However, there are on-going debates in the literature around whether we primarily feel real-life emotions, or rather aesthetic emotions, while listening to music (e.g. Eerola & Vuoskoski, 2012; Vempala & Russo; 2013). The so-called Cognitivists assume that musically evoked emotions are not comparable to those experienced in everyday life (Kivy, 1990); for example, they would argue that the sadness felt due to the loss of a close friend cannot be compared to the sadness felt after listening to a sad song. Scherer and colleagues have reported several studies of aesthetic emotions (see Scherer & Coutinho, 2013; Scherer & Zentner, 2001; 2008), which showed that these emotions are relatively specific to the intrinsic nature of music, or art more generally. Scherer & Coutinho

(2013) see support for this notion in studies that have explored the experience of bodily chills as a response to music. Given this type of experience does not appear to serve the same purpose as the experience of real-life emotions, this might suggest that aesthetic emotions are of primary importance while listening to music.

The Emotivists, on the other hand, suggest that music can evoke emotions comparable to those experienced in everyday life. This can be achieved by, for example, varying musical parameters (major = happiness, minor = sadness; compare Habibi & Damasio, 2014; Hunter et al., 2011; Mladjenović, Bogunović, Masnikosa & Radak, 2009; Schellenberg & Scheve, 2012; Shafron & Karno, 2013). One example that highlights the Emotivist perspective is the study by Ogg, Sears, Marin and McAdams (2017). This study suggests that physiological emotional responses while listening to music, assessed via electrodermal and cardiovascular measures and somatic muscle activity measures, are the same as real-life emotions. Neuroscientific approaches have also demonstrated that music, particularly an individual's favorite music, can produce brain activity in areas known to be associated with the experience of positive emotions (Blood & Zatorre, 2001; Montag, Reuter & Axmacher, 2011; see a review by Koelsch, 2011). Finally, Panksepp and Bernatzky (2002) have demonstrated that listening to sad music elevates sadness, while listening to happy music triggers a happy state of mind. Nevertheless, these two perspectives (e.g. emphasizing the importance of aesthetic emotions or real-life emotions) are not mutually exclusive, and the importance of both perspectives has been demonstrated in empirical studies (see Habibi & Damasio, 2014; Hunter et al., 2011; Juslin, 2013; Vempala & Russo; 2013).

Regardless of the debate around whether we primarily feel real-life emotions or aesthetic emotions while listening to music, the processes that trigger these emotions have not yet been fully elucidated. Recent studies have shown that empathy appears to be important in understanding individual differences in emotional reactions to music (Greenberg et al., 2015). This is in line with a theoretical account of this provided by Scherer and Zentner (2001). In their account, empathy is described as one of several factors involved in the production of musically conveyed emotions. Egermann & McAdams (2013) suggest that a similar concept to empathy, emotional contagion, may also be important in helping understand how emotional responses to music are produced. Unlike empathy, emotional contagion is characterized by a lack of self- and other-awareness. With empathic reactions, an individual can differentiate between their own and others' feelings; this is not the case, however, with emotional contagion (Egermann & McAdams, 2013). While empathy with a composer and unconscious emotional contagion while listening to music can be seen as related constructs (Egermann & McAdams, 2013), this study is only considering the role of empathy.

With regard to empirical work on empathy and music, Wöllner (2012) found that the perception of emotional expressiveness in music performance is related to empathy. The role of empathy has been further demonstrated in several studies which have shown evidence for an effect of dispositional empathy on the perception of musically conveyed emotion. Individuals with higher degrees of overall empathy were found to be more aware of, and were more affected by, the emotions induced via music, especially intense sadness, compared to less empathic people (e.g. Baltes & Miu, 2014; Greenberg et al., 2015; Kawakami & Katahira, 2015; Vuoskoski & Eerola, 2011; Vuoskoski & Eerola, 2012; Wöllner, 2012). As an example, a study by Eerola, Vuoskoski, and Kautiainen (2016) recently examined the phenomenon of liking sad music and found evidence for an association between trait empathy and sadness that is moving (i.e. the enjoyment of sad music). More specifically, the associations were found for the subscales Fantasy (FS), $r_{(99)} = .38$, and Empathic Concern (EC), $r_{(99)} = .35$ (both p < .001), from the Interpersonal Reactivity Index (IRI). Similar

findings have been observed in previous studies by Vuoskowski and Eerola (2011, 2012). The results of a study by Kawakami and Katahira (2015) also support an association between trait empathy and the enjoyment of sad music; in addition to the associations with EC and FS, they also found an association between the IRI-facet Perspective Taking (PT) and the enjoyment of sad music.

In these studies, the IRI, as one of the most widely used questionnaire measures of empathy, was chosen to assess trait empathy. More objective measures of empathy, such as physiological measures, which would allow further insight in to the mechanisms underpinning empathic ability are, however, still lacking (although there has been neuroimaging work on the mechanisms underpinning empathy in humans – see Decety, 2011, for an overview of this work). In addition to work using MRI to investigate empathy, a growing number of researchers have applied molecular genetics methods to examine empathy (e.g. McInnis, McQuaid, Matheson & Anisman, 2015; Melchers et al., 2013; Montag, Sindermann, Melchers, Jung, Luo, Becker et al., 2017). While studies of the heritability of empathy have shown large variation in the heritability estimate across study (between 0 and 70% heritability), perhaps due to the variety of measurement instruments and sample characteristics found in these studies, Melchers, Montag, Reuter, Spinath and Hahn (2016) showed heritability estimates between 52% and 57 % for affective empathy and a behavioral paradigm (Reading the Mind in the Eyes Test). For the cognitive measures of empathy, lower estimates were observed (27 %). These findings correspond with the results shown by Zahn-Waxler, Emde and Robinson (1992), who also found a stronger genetic influence on the affective component of empathy.

These heritability estimates are supported by studies that have revealed potential molecular genetic markers of empathy. Most notably, the oxytocin receptor gene was found to

be involved in explaining inter-individual differences in empathic abilities (e.g. Christ, Carlo & Stoltenberg, 2016; McInnis et al., 2015; Montag et al., 2017). Beyond oxytocin, other hormones have been proposed in the literature to be relevant for an understanding of empathy.

Huron (2011) suggested that prolactin could play an important role in musically conveyed emotions due to its association with empathic responses. More specifically, Huron suggested that individuals with higher prolactin levels should experience more pleasant responses to sad music, whereas individuals with lower prolactin levels should have more unpleasant emotional experiences (e.g. experience more sadness). This postulation attempts to explain the often paradoxical reactions of humans to sad music. Although sad music generally induces a sad mood (see Panksepp & Bernatzky, 2002), some individuals report positive emotions while listening to sad music (Juslin, 2013).

Prolactin, mainly released in the anterior pituitary, is well known in the context of reproduction and lactation (Freeman, Kanyicska, Lerant, & Nagy, 2000). It is also known to mediate the human affiliative bonding process (Neumann, 2009). The gene coding for prolactin (called PRL) has, however, unfortunately been under-studied compared to the neuropeptide oxytocin or the catecholamines. The PRL gene can be found on the chromosome 6p22.2- p21.3. Very little work examining the genetic associations between the PRL gene and phenotypes from social neuroscience can be found in the previous literature until quite recently. Most of the studies in the field so far have dealt with associations with breast cancer (Nyante et al., 2011; Vaclavicek et al., 2006). A few studies have also examined links between PRL or PRLR (coding for the prolactin receptor) and childhood mood disorders (Strauss et al., 2010). Given the current state of the literature, it is clear that the choice of single nucleotide polymorphisms (SNPs: variants of a single building block of DNA) should be exploratory and aim to test SNPs with clear observable variation in Caucasian populations

(i.e. avoiding ultra rare variants, which are hard to study given their rare occurrence in a given population, leading to a lack of power in statistical testing). On that basis, we decided to test for two SNPs of the PRL gene (rs1205960 with base pairs C/T and rs13354826 with base pairs A/G) in order to find evidence for the role of prolactin in music perception. Both SNPs are located in an intronic region of the PRL gene and therefore do not function in coding for protein synthesis. Hence potential associations, via influencing the functionality of the PRL gene, can only be established via linkages to SNPs in exonic or promoter regions (i.e. regions that are coding for protein synthesis).

The aims of the present study were to, firstly, examine how individual differences in empathy are related to affective and cognitive responses (i.e., felt and perceived emotions) after listening to happy and sad music. We predicted that after listening to the pieces of music, participants with higher scores on affective and cognitive empathy would show more congruent affective responses and cognitive evaluations, given the emotion that was conveyed through features of the composition (e.g. sadness by using the minor key and happiness by using the major key), (Baltes & Miu, 2014; Greenberg et al., 2015; Wöllner, 2012). Secondly, we sought to examine the potential association between the SNPs rs1205960 and rs13354826 of the PRL gene as possible markers of empathy and variables related to the perception and experience of emotions while listening to happy and sad music.

Materials and Method

Participants

Participants were recruited at Ulm University, Germany. A prerequisite for participation in the study was having contributed to the Ulm Gene Brain Behavior Project by the Department of Molecular Psychology, which required the completion of several questionnaires (including the IRI, among others) and DNA extraction from buccal cells. A total of 174 (N = 174) subjects participated in this study. Fourteen participants were excluded because of missing data (e.g., not fully completing the questionnaire). The final sample consisted of 160 participants ($n_{male} = 53$, $n_{female} = 107$), whose age ranged from 18 to 61 years (M = 24.77, SD = 8.16). Only 155 participants were included in the genetic analyses reported in this study because of failed genotyping in n = 5 participants. A total of 118 participants had learned at least one musical instrument, and 42 participants had not received any private music tuition. Most of the participants preferred listening to the pop genre of music (40.6%), followed by rock (30.0%), hiphop (9.4%) and classical music (6.3%). All participants gave their written informed consent before their participation in the study. The study was approved by the Ethics Committee of Ulm University, Ulm, Germany.

Genotyping

DNA was extracted from buccal cells. The purification was conducted with the MagNA Pure 96 system using the MagNa Pure 96 DNA kit from Roche Diagnostics, Mannheim, Germany. Genotyping of the PRL_rs1205960 (CC, CT, TT) and PRL_ rs13354826 polymorphism (AA, AG, GG) was implemented on a MALDI-TOF platform (Agena; Massarray 4) by Varionostic, Ulm, Germany. The distribution of the genotypes for each SNP across the sample is displayed in Table 1.

(Insert Table 1 about here)

Future studies should also consider examining other genes, including those coding for oxytocinergic neurotransmission. This might even be of interest in the field of Neuroeconomics, as suggested in a recent study (Riedl, Javor, Gefen, Felten & Reuter 2017). Given the work by Huron (2011), prolactin genetics was an obvious theoretical choice for the present study. As we have also analyzed three OXTR SNPs in our genetic bank for the participants in this study (and given oxytocin would also have been a good candidate gene approach for the present work), we provide non-significant findings for this data in the supplementary material as a reference for future research endeavors.

Self-Report-Measures

The IRI was used to assess empathy; this is one of the most frequently used questionnaire measures of empathy. The questionnaire was developed by Davis in 1980 as one of the first multidimensional approaches to the measurement of empathy. The cognitive and affective facets of empathy were assessed via four subscales, with seven items in each.

Cognitive empathy was represented by the subscales perspective taking (PT; the ability to look at something from the perspective of others; e.g.: "I sometimes try to understand my friends better by imagining how things look from their perspective") and fantasy (FS; the ability to engage with characters from novels and films; e.g.: "I really get involved with the feelings of the characters in a novel"). The affective component was measured with the subscales empathic concern (EC; sympathy with others; e.g.: "I would describe myself as a pretty soft-hearted person") and personal distress (PD; personal discomfort when confronted with the emotional state of others; e.g.: "I sometimes feel helpless when I am in the middle of a very emotional situation"). Responses were measured via a 5-point Likert-scale (0 = does not describe myself well; 4 = describes myself very well). Higher scores indicated higher PT, FS, EC and PD.

Assessment of the reliability and validity of the IRI support the use of this measure for the assessment of empathy. For example, test-retest reliability for women and men shows average values between rr = .61 and .81 (Davis, 1980). Internal consistency (Cronbach's α)

estimates for the IRI scales in the current sample, shown in Table 2, also support the use of this measure.

(Insert Table 2 about here)

The emotional responses of the participants were assessed with the Self-Assessment-Manikin (SAM) (Bradley & Lang, 1994). The SAM consists of three 9-point rating scales that measure valence, arousal and dominance. It is often used in research because it can be completed quickly and is simple to understand for participants. For example, a study of auditory stimuli and the SAM was conducted by Bradley (1994), where the SAM was shown to be a reliable instrument. However, both Morris and Boone (1998) and Zentner and Eerola (2010) suggest that the 9-point Likert scales measuring valence ("How do you feel?"; 1 = not good, 9 = very good) and arousal ("How excited are you?"; 1 = not excited, 9 = extremely excited) are sufficient for assessing emotional responses to music, so only these two scales were used in the current study.

Musical Stimuli

The song selection was based on a harmonic analysis of songs from different genres, such as pop / rock, jazz and classical music. The selection was mainly based on these genres, since they are the most popular and preferred genres in the general population (e.g. Deutsches Musikinformationszentrum, 2015). The final song selection consisted of 10 instrumental songs and 10 vocal songs, with five happy songs and five sad songs in each category (see Table 3).

(Insert Table 3 about here)

Every song was randomly presented for 30 s. Due to the balanced use of instrumental and vocal songs, the possible effects of emotions prompted by the lyrical content of the songs could be controlled. The musical analysis was based on the theory of Willimek and Willimek (2011), as well as on the criteria proposed by Eerola, Friberg and Bresin (2013) and Gabrielsson and Lindström (2010). Thus, the minor key and slow tempo were the main characteristics of sad songs. In contrast, the major key and fast tempo were indicators of happy music.

Procedure

The study was conducted as an online-experiment. Only 13 participants were tested in a standardized lab setting. These participants were included in the final sample because of the comparability of their data (i.e. there were no significant differences in t-tests comparing the online collected data with the data collected in the lab setting for all dependent variables). After the collection of demographic data, which included one item asking participants their most preferred music genre and another item assessing musical expertise, current emotional state was measured as a baseline (via the SAM). This was followed by the random presentation of the 20 sad and happy song excerpts for 30 seconds each at a pre-set volume of 75 dB, although it should be noted that the volume level cannot be verified in online experiments. After each song, the SAM scales for arousal ("How excited do you feel after listening to the song?" 1 = not excited, 9 = extremely excited) and valence ("How good do you feel after listening to the song?" 1 = not good, 9 = very good) were presented to assess the affective responses. After this, the participant's prior awareness of that particular song was measured ("Did you know the song you have just heard?"), as well as existing positive, negative or neutral memories associated with that song ("Do you have any memories of the song you have just heard? If so, were they positive, negative or neutral?"). After this, the

cognitive evaluation of the emotional song content was assessed via two self-constructed scales ("How sad/cheerful would you rate the emotional content of this song (your rational evaluation, not what you felt)?; 1 = very sad / not at all cheerful to 11 = very cheerful / not at all sad. For the sake of clarity, the scales were inverted to 1 = not sad / happy to 11 = very happy / sad in the results section. Figure 1 displays a graphical overview of the procedure that followed each song excerpt.

(Insert Figure 1 about here)

We should draw attention to the distinction made between evoked or induced affect as a response to the music (i.e., felt emotions), and an evaluation of the affect that has been presented in the music (i.e., perceived emotions). This distinction is reflected in the valence and arousal questions (which were posed immediately following the musical stimuli) and the sadness and happiness questions (which were posed after the items asking about existing memories of the song) described in the scales above. The items for valence and arousal targeted the induced (felt) affect, in contrast to the happiness and sadness ratings that were designed to assess the represented (perceived) affect.

Statistical analysis

Statistical analysis was conducted using *IBM SPSS Statistics 21*. Since we were interested in the emotional responses after listening to sad and happy songs in general, and not emotional reactions to specific songs, we calculated the arithmetic mean over the ratings in response to all ten songs of each emotional category. Using this procedure, we produced eight variables that represented the perceived emotions (four variables: rating of sadness and

happiness for both emotional qualities) and the felt emotions (four variables: arousal after happy/sad songs, valence after happy/sad song).

The first hypothesis, positing a relationship between the subscales of the IRI and the felt/perceived emotions, was tested by calculating *Pearson correlation coefficients*, as well as *partial correlations* (controlling for the felt emotions in terms of the perceived emotions, and vice versa). The second hypothesis, positing an association between the PRL gene and the felt/perceived emotions, was tested by calculating two separate *ANOVAs*. Each *ANOVA* had one SNP (i.e. polymorphisms of the PRL gene) as a fixed variable (PRL_rs1205960 with the genotypes CC, TC, TT; PRL_rs13354826 with the genotypes AA, AG, GG).

Results

Descriptive Analysis

Descriptive statistics for the perceived emotions (ratings of the displayed sadness / happiness of the songs) and felt emotions are listed in Table 4 for sad songs and for happy songs. Average values for the perceived emotion variables show satisfactory estimation of the target emotion conveyed in the music, but the range of the ratings indicate there are inter-individual differences in these ratings.

(Insert Table 4 about here)

Manipulation Check

Paired samples *t-tests* were conducted as a manipulation check in order to examine if the participants evaluated the emotional content of the songs as predicted; that is, we assessed if the sad songs were categorized as sad, and if the happy songs were characterized as happy. The results of these analyses support this proposition. More specifically, the evaluation of perceived sadness significantly differed while listening to sad songs (M = 6.68, SE = 0.09) and happy songs (M = 1.63, SE = 0.10, with higher sadness ratings for sad songs than for happy songs (t(318) = 39.00, SE = .13, p < .001). Also, evaluation of perceived happiness significantly differed while listening to sad songs (M = 3.87, SE = 0.09) and happy songs (M= 9.05, SE = 0.08), with higher happiness ratings for happy songs than for sad songs (t(318) =42.54, SE = 0.12, p < .001).

Effect of PRL SNPs rs1205960 and rs13354826 on Empathy (IRI)

Another prerequisite was checking the effect of the two SNPs rs1205960 and rs13354826 of the PRL gene on empathy. A *MANOVA* showed no significant effects of both SNPs on the four IRI subscales, as well as no significant interaction between rs1205960 and rs13354826.

Analyses of possible confounding variables

To examine possible confounding variables like genre preference (rock, pop, classic, r`n`b, hip-hop, jazz, electro, folk) and musical expertise (0 = Yes, 1 = No), *Spearman correlation coefficients* for genre preference, as well as *point biserial correlation coefficients* for musical expertise, were computed. The *spearman correlation test* of genre preference on the perceived/felt emotion was not significant. Significant, but small, correlations were found for musical expertise and felt emotions (valence) in both conditions (happy: $r_{(158)} = -.25$, p < .01; sad: $r_{(158)} = -.21$, p < .01), for the perceived sadness level of sad songs ($r_{(158)} = -.20$, p < .05) and of happy songs ($r_{(158)} = .18$, p < .05) and for the perceived happiness level of happy songs ($r_{(158)} = -.16$, p < .05.). However, it should be noted that these relatively small correlations potentially diminish existing effects due to increased SEs and thus a decrease in power (see Kahan, Jairath, Doré & Morris, 2014, Spector & Brannick, 2011). Consequently,

musical expertise was not considered to have any undue influence here. This assumption is confirmed by the fact that our key results were still largely found after controlling for musical expertise. Empathy has previously been shown to be confounded with gender. Females often report higher empathy scores and typically perform better on socio-emotional tasks (Derntl et al., 2010). Since the results of the *ANOVA* including gender did not result in a significant main effect of gender, nor in a significant interaction effect (Gender*SNP), gender was not considered to have any undue influence on the results of the study.

IRI

The correlation between the IRI subscales and the perceived/felt emotions while listening to the music excerpts revealed a significant negative correlation between PD and the valence rating of happy songs ($r_{(158)} = -.20$, p < .05) and sad songs ($r_{(158)} = -.21$, p < .01). PD was also negatively associated with the evaluation of the perceived happiness ratings of sad songs ($r_{(158)} = -.17$, p < .05). For FS there was a significant negative correlation between the evaluation of the perceived happiness of sad songs ($r_{(158)} = -.22$, p < .01) and a positive correlation between FS and the perceived sadness evaluation ($r_{(158)} = .21$, p < .01) of sad songs. EC was associated with the perceived sadness evaluation of sad songs ($r_{(158)} = .22$, p <.01) and with the happiness evaluation ($r_{(158)} = -.17$, p < .05) of sad songs. No other significant correlations were found. It should be noted that all but one (PD and the evaluation of perceived happiness of sad songs) of the significant associations described above were also found using *partial correlations* controlling for felt emotions (in terms of associations between IRI scales and perceived emotions), and perceived emotions (in terms of associations between IRI scales and felt emotions), respectively. These results are shown in Table 5.

(Insert Tables 5)

Associations between genetic variations of the PRL gene and felt/perceived emotions

Before the hypotheses were tested, some initial checks on the data were conducted. Firstly, baseline measures from the SAM (i.e. felt emotions) were tested to exclude existing differences between the genotypes prior to listening to the music excerpts. Given the results of a *MANOVA* showed no differences in terms of the PRL_1205960, we did not adjust the subsequent analyses for this SNP. Secondly, we performed the *Hardy-Weinberg equilibrium* (*HWE*) test to rule out any abnormal distribution of the PRL_rs1205960. This SNP distribution did not deviate from *HWE*: $X^2(1) = 0.12$, p = .73 (CC = 86, CT = 60, TT = 9). Furthermore, we checked differences across other relevant variables. Gender ($X^2(2) = .70$, p = .71), age (F(2,152) = 0.05, p = .95) and musical expertise ($X^2(2) = 1.51$, p = .47), were equally distributed across the genotypes.

The *ANOVA* with PRL_rs1205960 as a fixed factor only showed significant effects for arousal (as one indicator of felt emotions). While listening to sad songs, CC carriers (M = 3.85, SE = 0.09) reported higher arousal than CT- (M = 3.35, SE = 0.10) and TT carriers (M = 2.97, SE = 0.10). The overall significant effect (F(2,152) = 4.41, p = .014, $\eta^2 = .06$) was then further analyzed using post hoc tests in relation to specific genotype comparisons (Bonferroni correction: $\alpha = 0.5/4$ resulting in .0125; two SNPs, plus the test for arousal and valence); TT vs. TC, CC vs. TT, TC vs. CC). For sad songs, the CC- and CT carriers differed significantly, but both groups did not differ in comparison with TT carriers.

The genotype groups also differed significantly in response to the happy songs. Here, the same pattern was found as for sad songs. CC carriers (M = 4.28, SE = 0.09) had higher

ratings on arousal than CT- (M = 3.69, SE = 0.12) and TT carriers (M = 2.96, SE = 0.09), F(2,152) = 5.61, p = .004, $\eta^2 = .07$). Post-hoc analysis showed significant differences between all three genotypes. CC- and CT carriers differed in their mean arousal ratings, and CC- and TT carriers also differed. Aside from the effect of variations of the PRL_rs1205960 for arousal, no other significant effects were found. Participants did not differ on their perceived emotions and valence ratings.

Following the analyses above, a second set of analyses was undertaken. TC- and TT carriers were combined in to one group due to the small number of TT carriers (n = 9). Two ttests were then separately conducted for perceived/felt emotions as outcome variables, with the genotype groups of the PRL_rs1205960 as a fixed variable (CC vs. TC/TT). For the aggregated genotypes (CC vs. CT/TT), the *t-tests* revealed a significant effect for arousal level in both emotional categories (see Figures 2 and 3). For sad songs, the genotypes significantly differed (t(153) = 2.84, p = .005, Cohen's d = .46), with the CT/TT carriers showing less arousal (M = 3.30, SE = 0.10) than CC carriers (M = 3.85, SE = 0.09). The same effect could be seen for arousal level after listening to happy songs. The two groups significantly differed (t(153) = 3.00, p = .003, Cohen's d = .48), with lower arousal levels in the CT/TT group (M = 3.60, SE = 0.12) compared to the CC group (M = 4.28, SE = 0.11). With regards to valence, the values of CC carriers and CT/TT carriers did not significantly differ (happy songs: t(153)) = 0.30, p = .77, sad songs: t(153) = -0.36, p = .73). Similar effects were obtained for the perceived emotion variables. As one can see, the effect for PRL_rs1205960 on allele levels holds when correcting for multiple testing. The effect for arousal by genotype level after the happy songs passed these correction procedures, while for arousal after the sad songs, the threshold was only just missed (compare .014 to .0125).

(Insert Figure 2 and 3 about here)

For the second SNP of the PRL gene (rs13354826), a similar set of analyses was undertaken. Before testing the hypothesis, we ruled out differences for the baseline measure of the *SAM* scales. Since the baseline measure (specifically the valence baseline) differed significantly for variants on the PRL_rs13354826, we adjusted the ratings of felt emotions for this factor in the test of the hypothesis. The distribution of this SNP did not deviate from $HWE(X^2(1) = 0.83, p = .36, AA = 62, AG = 66, GG = 24)$. Gender ($X^2(2) = 0.32, p = .86$), age (F(2,149) = 0.52, p = .60) and musical expertise ($X^2(2) = 1.36, p = .51$), were equally distributed across the genotypes.

The results of the *ANOVAs* run for testing the hypothesis on PRL_ rs13354826 revealed no significant effects for either perceived emotions, nor felt emotions. The p-values varied between .2 and .9 for all variables. An analysis of possible interactions between the two SNPs also revealed no significant effects. Aggregation of the rare genotype group with the heterozygote group was not undertaken given the reasonable number of GG carriers (n = 24). The main results of the PRL gene association analyses for both gene sections are presented in Table 6.

(Insert Table 6 about here)

Discussion

The aim of the present research was to, firstly, investigate how empathic traits are associated with the emotional perception/experience of sad and happy music. Secondly, we were interested in examining if prolactin, as a possible biological marker of empathy, plays an important role in understanding the effects of music on the listener. Despite an association between the PRL gene and trait empathy not being found, we suggest that the PRL gene is involved in the experience of emotions while listening to music, particularly in relation to levels of arousal in response to music.

IRI

In relation to the IRI, significant positive correlations were found between FS and EC and the emotional perception of sad songs. Participants with higher scores on these subscales (hence higher empathy) evaluated sad songs more accurately than participants with lower scores. Also, participants with higher scores on PD evaluated the happiness of sad songs more accurately than participants with lower PD. Significant correlations with the felt emotions were only found for the PD subscale. Higher values on PD were associated with higher negative ratings of valence for happy and sad music. On that basis PD, which is a selfdirected aversive state when confronted with another's distress (Eisenberg & Strayer, 1990; example items include "I sometimes feel helpless when I am in the middle of a very emotional situation" (high PD) and "When I see someone get hurt, I tend to remain calm" (low PD)), seems to facilitate empathic reactions when being exposed to sad music, whereas it hinders empathic reactions to happy music. This is in line with the previous literature which suggests that people with higher values on PD are not only negatively affected by being confronted with the aversive states of others, but also more easily relate to the distressing situations and feelings of others (see Konrath & Grynberg, 2013). Konrath and Grynberg (2013) explained this as follows: "Another way to think of this is that in order to be truly empathic, people will indeed suffer with those who are suffering (and thus feel some distress on behalf of them). (...) [Therefore] Personal distress often includes unmitigated contagion with the suffering person, along with over-identification and poor personal boundaries." (p.8). An alternative explanation could be that people scoring higher on PD tend to evaluate things

more negatively in general, since PD was found to be positively associated with neuroticism (e.g. r = .55; p < .001, in a study by Melchers, Li, Haas, Reuter, Bischoff & Montag, 2016).

The associations of FS (empathizing with fictional characters) and EC (trying to fulfill another's needs, or attenuating their distress) with perceived emotions while listening to sad songs indicates that people with higher empathic abilities in these two domains are more accurate in their cognitive evaluation of the emotional quality of the music, whereas these subscales did not relate to the affective responses (i.e. felt emotions). Aside from the correlation of PD with the valence rating for both emotional categories, no association between self-reported empathy and felt emotions while listening to the musical excerpts was found.

One possible explanation for the lack of associations between felt emotions and empathy could be that people enjoy listening to sad music (see also Eerola et al., 2016). Thus, people may not respond emotionally in the intended direction. This phenomenon might be especially true for empathic people (Eerola et al., 2016), which might explain the nonsignificant relations found in the current study.

The relationship found between cognitive empathy (concerning the ability to empathize with the feelings of fictional characters; FS) and cognitive emotional evaluation (i.e., perceived emotions) suggests that this part of trait empathy is more relevant for the cognitive processing of musically evoked sadness, but may not be as relevant for affective processing of music. This may account for the lack of a relationship between empathy and felt emotions while listening to music in this study. It is, however, important to note that previous studies have shown associations between trait empathy (especially FS and EC) and emotional reactivity to music (e.g., Eerola et al., 2016; Vuoskoski & Eerola, 2012). Another point to note is that we mostly found correlations for the emotional perception of sad songs (except in relation to PD, which should be treated as a unique facet of empathy). In non-musical settings, the influence of specific emotional qualities on empathic abilities has previously been shown (Olderbak, Sassenrath, Keller & Wilhelm, 2014). For music, the results might suggest a moderating effect of the quality of the emotion on the relation between empathy (especially FS and EC) and perceived emotions while listening to music, with research showing a stronger involvement of empathy while listening to sad songs specifically. This suggests that people scoring higher on the facets FS and EC are more accurate in their evaluation of the emotional content of sad songs, whereas individuals do not differ, or perform more poorly (PD), in their evaluation of happy songs as a function of their empathic abilities. In support of this, Greenberg et al. (2015) showed that perceptions of sad music were more dependent on empathic ability than those of happy music.

PRL

Analysis of the role of the PRL gene on Empathy revealed no significant effects of the two SNPs rs120596 and rs13354826 across all subscales. The PRL gene was not associated with either cognitive or affective empathy, even though previous studies have shown effects of the PRL gene on empathy-related factors such as emotional bonding (e.g. Neumann, 2009). Previous research has shown associations between the PRL gene and the regulation of emotions (Turner et al., 2002) and maternal care (Pedersen, 2004). The lack of effect of the PRL gene on trait empathy shown in the current study could potentially be explained by, firstly, noting that the IRI may not be the most appropriate measure to use when investigating the genetic bases of empathy because it may represent a more environmentally shaped form of empathy, as suggested by Bischof-Köhler (1991). The heritability of this measure might perhaps suggests otherwise though (Melchers et al., 2016).

Secondly, there has been limited research on the PRL gene, consequently little is known about possible genetic markers of constructs like emotional bonding (Neumann, 2009) that have been previously associated with the PRL gene. More specifically, no previous study has been conducted on PRL gene associations with emotional responses while listening to music in an attempt to give the theoretical work of Huron (2011) empirical support. This work provided the main theoretical backdrop to the exploratory postulation in this study that variations on the prolactin gene would affect musically induced emotions.

Thirdly, it is possible that other combinations of SNPs of the PRL gene are related to inter-individual differences in empathy. Lastly, the PRL gene, aside from its associations with empathy related constructs, may not be related directly to empathy per se, but rather with inter-individual PRL hormone levels (which we could not measure over SNP analyses). The effects of the PRL hormone on, for example, affective bonding (e.g. Neumann, 2009) that have previously been shown might therefore suggest environmentally influenced (perhaps epigenetic) effects. These points should be clarified further in experimental studies.

Nevertheless, the current study showed a significant effect of the rs1205960 on arousal, as an indicator of an affective response (i.e., felt emotion), while listening to music. C carriers on the rs1205960 showed a stronger physiological response than T carriers; the rs13354826 did not show any effects on either perceived or felt emotions. This link between genetic variations on the PRL_rs1205960 and felt emotions while listening to music should be clarified in further research. The effect of rs1205960 on arousal is potentially interesting because no correlations between arousal and the subscales of the IRI were found. This may be an indication of a stronger effect of genetically-influenced systems that might be associated with empathy related constructs on more physiologically-based affective reactions, whereas perceived emotions depend more on trait empathy that is developed over the life-span, with stronger influences from culture and personality (Bischof-Köhler, 1991). A stronger genetic influence for affective empathy has already been found in heritability studies (e.g. Melchers et al., 2016; Zahn-Waxler et al., 1992). Therefore, our results suggest that a bottom-up effect of genetically-influenced systems likely affects emotional circuits in our brain (Panksepp, 1998) and involves the activity of specific hormones/neurotransmitters.

Following Huron's (2011) theoretical suggestion, higher levels of prolactin might be responsible for the feeling of pleasure individuals experience while listening to sad music. His theory presumes an effect of prolactin analogous to the effect of endorphins. It suggests the release of prolactin protects people from feeling grief and distress while listening to sad music. Variations of the PRL gene have already been associated with the functioning of the PRL system and emotional functioning (Hirata, Zai, Nowrouzi, Shaikh, Kennedy & Beitchman, 2016). Considering Huron's theory of the emotional response to sad music, the results of this study support the putative association between prolactin and emotional functioning (specifically on arousal). The differences found between T- and C carriers in their physiological emotional reactions while listening to sad and happy songs can be interpreted in line with Huron's theory of an hedonic effect of prolactin. T carriers showed less arousal towards sad and happy songs. This might be regarded as indicative of an association between prolactin levels and felt emotions while listening to music (clearly our findings do not yield evidence for an effect of the SNPs studied here, or polymorphisms in linkage, on prolactin levels).

The lack of an effect of the PRL_rs1205960 on the valence rating and on the perceived emotions might suggest that even if the levels of felt arousal are reduced in T carriers, the rating accuracy of the perceived emotion is not affected. Therefore, one might assume that differences in the genetic make-up of the prolactin system do not influence social perception

in the way studies have shown for the oxytocin system, but that they probably influence physiological susceptibility while being exposed to highly emotional stimuli (McInnis et al., 2015).

The knowledge that empathy, namely FS & EC, can be relevant for the perception of emotions, especially for sad songs, is of great importance in a range of contexts, for example the training of musicians. Heisel (2015) has already suggested that more empathic musicians can convey emotions more clearly and convincingly, which is often of great importance for the success of their performance. The results found here can be interpreted as support for this suggestion, meaning that some facets of empathy may be essential to consider in the apprenticeship of musicians.

The link between empathy and the perception of emotion in music is also important for therapeutic approaches based on music. ASD treatments often target the cognitive empathic abilities of the patients, since they show massive deficits in this respect (Panksepp & Bernatzky, 2002). If empathy is essential for the accurate perception of emotions evoked by music, then focused musical treatment could help people with deficits in cognitive empathy to learn empathic responses (e.g. randomized controlled trials (RCTs) have already shown significant effects in music therapy interventions for children with ASD; see Gold, Wigram & Elefant, 2006). Patients would thus profit from musical interventions since music supports intrinsic motivation, social competence and well-being (e.g. Nilsson, 2009; Panksepp & Bernatzky, 2002).

Limitations

There were some limitations in the design of this study. Firstly, it should be noted that the emotional responses to the music were only assessed via the SAM. For a better

understanding of the effects found in the study, additional measurement of physiological parameters, such as heart rate or skin conductance, is essential. This also applies to the genetic hypotheses; our results should be replicated with additional hormone level analyses for a deeper understanding of the association found between the prolactin transmitter system and musically induced emotions.

Secondly, it should be noted that we found an effect of the aggregated genotype of PRL_rs1205960 (CC; CT/TT) on baseline arousal (p = .04). Given that this effect was not found at the genotype level (i.e. CC vs. CT vs. TT), we did not control for this in the analyses reported in the result section. Nevertheless, it is worth highlighting that when including baseline arousal as a covariate in our analyses, we still found a stable effect of PRL_rs1205960 (CC vs. CT/TT) on arousal after listening to happy songs (F(1,152) = 4.58, p = .03) but barely not for arousal after listening to sad songs (F(1,152) = 3.70, p = .056). This indicates that the PRL rs1205960 effect on arousal after listening to happy songs represents the more robust effect.

Finally, emotional reactions, especially while listening to music, depend on several factors, such as being alone or in a familiar environment, the loudness and intensity of the sound, familiarity and preference for the music, and emotional state while listening to the music (e.g. Korsakova-Kreyn & Dowling, 2014, Taylor & Friedman, 2014). In this study, some factors that might influence emotional experiences while listening to music were controlled, for example genre preference. Due to the online nature of the study, the participants also had the opportunity to listen to the music in a private, familiar environment. Nevertheless, loudness and intensity depend on the quality of the speakers used by participants, which could be not controlled in this study. The duration of the musical excerpts may also have influenced the emotional response. It is possible that 30 s is not long enough to

evoke strong, conscious emotions. Another factor that may have influenced the results was the total number of musical excerpts presented; 20 musical excerpts may have induced fatigue in the participants. The musical excerpts used in this study were chosen on the basis that they would not be unduly influenced by genre and vocal style. Nonetheless, the relatively large number of musical excerpts, combined with their short duration, might have reduced the emotional impact of the pieces. Finally, the association between music, emotions and empathy has mostly been investigated using correlational or quasi-experimental designs. Experimental studies should be undertaken to strengthen the links found here between some facets of trait empathy and felt/perceived emotions while listening to music. A causal examination of these links is important, because deeper knowledge on this topic could also lead to a better understanding of empathy-related disorders like ASD, and could help support affordable and efficacious therapeutic approaches.

Ethical approval

Ethical approval for this project was given by the Ethic Committee of Ulm University.

Contributions

MS and CM designed the study. MS conducted the study (data collection), carried out statistical analysis and drafted the first version of manuscript. CM critically revised the manuscript. AC worked over the complete manuscript again, made critical amendments and improved the language.

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Citations in Supplementary Material

Baron-Cohen, Hoekstra, Knickmeyer & Wheelwright (2006) appears only in the supplementary material.

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Figures



Figure 1. Flowchart of the experimental procedure after listening to each song excerpt.



Figure 2. Average arousal rating after listening to happy songs (+/- 1 SE).



Figure 3. Average arousal rating after listening to sad songs (+/- 1 SE).

Table 1

Genotype distribution

		PRL_1205960		PRL_13354826				
Genotype	CC	СТ	TT	AA	AG	GG		
N	86	60	9	62	66	24		
%	55.5	38.7	5.8	40.8	43.4	15.8		

Note. $N_{PRL_{1205960}} = 155$. $N_{PRL_{13354826}} = 152$.

Table 2

Internal consistency (Cronbach's α) for IRI subscales

IRI scales	FS	EC	PD	РТ	
Cronbach's a	.86	.83	.79	.84	

Note. Seven items per subscale. FS = Fantasy, EC = Empathic Concern, PD = Personal

Distress, PT = Perspective Taking.

Song list for each emotional category

	Sad songs		Happy songs					
Artist / Composer	Titel	Instrumental/Vocal	Artist/Composer	Titel	Instrumental/Vocal			
Angele Dubeau	Nocturne op. 20 in C sharp	Instrumental	London Chamber Orchestra	Harry Potter & the	Instrumental			
(Violin) & La Pieta	minor, op. posth.		/ N. Hooper	Halfblood Prince				
(Piano) / F. Chopin				Soundtrack – Fireworks				
J. Williams	Schindler`s List	Instrumental	Vienna Philharmonic	Tritsch-Trasch-Polka	Instrumental			
	Soundtrack – Main Theme		Orchestra / J. Strauss					
J. Horner	Titanic Soundtrack – Main	Instrumental	Budapest Scoring	Can Can	Instrumental			
	Theme		Symphonic Orchestra / J.					
			Offenbach					
Yiruma	River flows in you	Instrumental	S. Joplin	The entertainer	Instrumental			

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London Chamber	Harry Potter & the	Instrumental	G. Bizet	Carmen - Ouverture	Instrumental						
Orchestra / N. Hooper	Halfblood Prince										
	Soundtrack – Dumbledores	undtrack – Dumbledores									
	Farewell										
Queen	My Melancholy Blues	Vocal	Taylor Swift	Shake it off	Vocal						
Christina Aguilera	Hurt	Vocal	Mika	Lollipop	Vocal						
A great big world &	Say something	Vocal	The Beatles	Twist and Shout	Vocal						
Christina Aguilera											
Sarah Connor	From Sarah with love	Vocal	Edgar Ott & Stefan	Probier`s mal mit	Vocal						
			Sczodrok	Gemütlichkeit							
C. Dion	My way	Vocal	Roger Cicero	Die Liste	Vocal						

Descriptive data	of the felt and	perceived	emotional	responses
1	5 5	1		1

		Sad	songs	Нарру	/ songs
		M SE		М	SE
Perceived	Happiness	3.87	0.09	9.05	0.08
emotions	Sadness	6.68	0.10	1.63	0.10
Felt	Arousal	3.59	0.10	3.95	0.11
emotions	Valence	5.23	0.10	6.71	0.08

Note. N = 160. Perceived emotions: Happiness = Evaluation of happiness (1 = not happy, 11 = very happy) Sadness = Evaluation of Sadness (1 = not sad, 11 = very sad. Felt emotions: SAM scales for arousal and valence (Valence: 0 = unhappy, 9 = very happy; Arousal: 0 = not excited, 9 = very excited).

Correlation coefficients (r) between IRI subscales and perceived and felt emotions

		Happy Songs					Sad songs				
		Felt emotions		Perceived emotions		Felt emotions		Perceived e	motions		
		Arousal	Valence	Happiness	Sadness	Arousal	Valence	Happiness	Sadness		
Cognitive	РТ	03	00	.04	.07	.05	.10	04	.11		
empathy	FS	02	02	.04	05	.06	.06	23**	.20*		
Affective	EC	11	01	.07	09	04	.12	21**	.25**		
empathy	PD	.06	19*	07	19*	.10	16 ^a	15	.09		

Note. N = 160. Correlations coefficients were tested controlling the felt emotions for the perceived emotions, and the perceived emotions for the felt emotions. Perceived emotions: Happiness = Evaluation of happiness (1 = not happy, 11 = very happy), Sadness = Evaluation of Sadness (1 = not sad, 11 = very sad). Felt emotions (Valence: 0 = not good, 9 = very good, Arousal: 0 = not at all excited, 9 = very excited). PT = Perspective Taking, PD = Personal Distress, FS = Fantasy, EC = Empathic Concern. *p < .05, **p < .01. *p = .05.

		PRL_1205960			PRL_1205960_combined				PRL_13354826				
		Sad songs		Happy Songs		Sad songs		Happy Songs		Sad songs		Happy Son	
		F	р	F	р	F	р	F	р	F	р	F	р
Perceived emotions	Happiness	0.57	.57	0.27	.76	0.31	.58	0.51	.48	0.23	.80	0.63	.53
emotions	Sadness	0.23	.79	0.06	.94	0.34	.56	0.03	.87	0.17	.84	0.79	.46
Felt emotions	Arousal	4.41	.01	5.61	.00	8.03	.01	8.97	.00	0.21	.82	0.40	.67
emotions	Valence	0.09	.92	0.05	.95	0.13	.72	0.09	.77	0.25	.78	0.38	.68
	155 N		150 D	DI 100			ז תת וידי	12050	<u> </u>			וחח	122540

Main results of the ANOVAs calculated for PRL_1205960, PRL_1205960_combined and PRL_13354826

Note. N_{PRL_1205960} = 155. N_{PRL_13354826} = 152. PRL_1205960 (CC, TC, TT), PRL_1205960_combined (CC, CT/TT), PRL_13354826

(AA, AG, GG). Perceived emotions: Happiness = Evaluation of happiness (1 = not happy, 11 = very happy) Sadness = Evaluation of Sadness (1 = not sad, 11 = very sad. Felt emotions: *SAM* scales for arousal and valence (Valence: 0 = unhappy, 9 = very happy; Arousal: 0 = not excited, 9 = very excited). *df* PRL_13354826 (2,148). *df* PRL_1205960 (1,152). *df* PRL_1205960_combined (1,152).

Supplementary Material - Integral