



Attachment style and emotion regulation: the modulatory role of A118G polymorphism

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Abstract

Emotion regulation (ER) refers to the process by which one can regulate his/her own and other's emotions and is pivotal for both social functioning and emotional well-being. During infancy, the attachment relationship serves as a context within which a child develops emotional and social abilities. The tight link between attachment and emotion regulation is sustained by evidence showing that the diverse attachment styles are associated with different emotion regulation strategies. Despite this relation has been widely reported, the biological modulatory factors of such a link are still unknown. In this context, one potential candidate is the the μ -opioid receptor 1 gene (OPRM1). The A118G single nucleotide polymorphism (snp) in this gene has been indeed described as modulating the sensitivity to social rejection and the capability of experiencing pleasure from social and affective relationships. Here, we hypothesize that the A118G snp modulates the impact of the attachment style on emotion regulation.

Methods. We investigated this issue in a sample of 87 young adult individuals by a pilot study. Within this group, attachment style and ER were respectively evaluated by the Relationship Questionnaire and the Difficulties in Emotion Regulation Scale. Saliva DNA samples were collected and then genotyped for A118G snp. **Results.** A118G snp modulates the effect of the attachment style on ER. Specifically, G-allele carriers with insecure attachment showed significantly higher DERS total and Goals scores. This effect was absent in the A-allele carriers. G-allele carriers with insecure attachment showed the highest value among the four groups, whereas the G-allele carriers with secure attachment showed the lowest value among them in all these scales. **Conclusions.** This pilot study offers new insight on the factors that can shape emotional development, shedding light on the putative role of the opioid system in modulating the developmental trajectories of ER in relation to the attachment style.

Keywords: attachment style; emotion regulation; A118G polymorphism; clinical psychology.

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Introduction

Emotion regulation has important implications for social functioning and emotional well-being (Thompson et al., 2019; Livingstone & Srivastava, 2012; Aldao et al., 2010), and includes a series of strategies and behaviors implemented by the individual to regulate the emotion felt at a given moment (Thompson et al., 2019; Aldao et al., 2014; Goldin et al., 2008). So, being able to flexibly regulate one's emotions is critical for adaptive functioning across the life span (Helion et al., 2019). Several aspects of emotion regulation have been associated with positive developmental outcomes (e.g., social competence; Contreras et al., 2000). In parallel, emotion dysregulation has been described as a transdiagnostic risk factor involved in a large number of psychopathologies such as eating disorders and mood disorders (Prefit et al., 2019; Thompson et al., 2019).

The attachment relationship serves as a context within which a child develops emotional and social abilities (Cooke et al., 2019). This primary relationship has a safety-regulatory function that prompts the child to seek support from the parents in situations promoting negative affect. On the other hand, children's ability to use an attachment figure as a secure base provides the self-regulatory capacities essential to explore and manage everyday situations (Waters & Cummings, 2000). Several reports described a relation between the attachment system and emotion regulation, highlighting how the different attachment styles can differentially affect emotion regulation (Cooke et al., 2019; Mikulincer & Shaver, 2019). Secure-attached individuals are more likely to manage stressful situations by employing successful coping and emotional regulation strategies. These subjects are confident of the emotional availability of the attachment figure to provide support in times of distress and need. Moreover, they are open to their-own emotional inner world and are fully able to experience, express, and communicate emotional states (Bowlby, 1988). By contrast, secondary attachment strategies (Main, 1990) - namely attachment anxiety and attachment-related avoidance - are known as risk factors for difficulties in emotion regulation (Mikulincer & Shaver, 2016a; 2003). Subjects with anxious attachment use *hyperactivating strategies* to force a relationship with a significant other who is perceived as not sufficiently available and responsive to their attachment needs (Mikulincer & Shaver, 2019; Cassidy & Berlin, 1994). On the contrary, *deactivating strategies* are typically observed in avoidant-attached individuals. These strategies are characterized by the inhibition of support seeking and commitment as well as by the tendency to handling distress autonomously, with the aim of preventing the frustration caused by rejection by significant others (Fraley & Shaver, 2000; Main, 1990). These polarized emotion regulation strategies result from early attachment experiences with parents who were perceived as unable to meet the child's basic needs for care and affection (Mikulincer & Shaver, 2016b; Cassidy, 1994; Main, 1990).

Despite the tight link between attachment system and emotion regulation has been widely reported, the biological modulatory factors of such a link are at the moment unknown. Within this context, one candidate that urgently needs to be explored is the μ -opioid endogenous receptors (MOR) system.

According to "*The Brain Opioid Theory of Social Attachment*" (Panksepp et al., 1978) social contact relieves from discomfort and induces positive emotions through the release of endogenous opioids, while the social isolation causes opioid withdrawal symptoms and negative affect. Given the above, the motivation to establish and maintain social and affective bonds would depend on the balance between endorphins release and the withdrawal (Panksepp et al., 1978; 1980). Several studies have shown the involvement of the MOR system in the modulation of pain and pleasure (Leknes & Tracey, 2008), food seeking behaviors (Berridge, 1996), and hedonistic pleasure, such as the one for sex (Mahler & Berridge, 2011) or affiliation (Lutz & Kieffer, 2013).

Specifically, the μ -opioid receptor 1 gene (OPRM1) is a key gene of the MOR system, and its A118G single nucleotide polymorphism (snp) has been largely studied in psychological research context (Cimino et al., 2020; Collins et al., 2018; Troisi et al., 2012; Troisi et al., 2011; Krosiak et al., 2007). This polymorphism, among those of the opioid receptors, is the only one affecting the MOR density in the brain, with the G allele variant displaying a lower MOR availability compared to that shown by A allele variant (Peciña et al., 2015; Krosiak et al., 2007). The A118G snp has been reported to modulate individual differences in the ability to experience social reward. Specifically, the G-allele has been defined as "gain-of-function" variant, since G-carriers seem to benefit more from social affiliation than A-carriers, showing an increase in positive emotions/affectivity, and are more negatively affected by social isolation, showing increased negative emotions/affectivity (Higham et al., 2011; Troisi et al., 2011; Way et al., 2009; Barr et al., 2008). In particular, it was reported that G-carriers exhibit dispositional sensitivity to social rejection (Way et al., 2009) and a great tendency to experience more pleasure from social and affective relationships (both in healthy and in clinical context), with respect to A-carriers (Troisi et al., 2011).

In light of this evidence, we hypothesize that the A118G snp modulates the impact of the attachment style on emotion regulation. Here we investigated this issue by a pilot study in a sample of young adults.

Materials and Methods

Participants

Participants included a group of 87 volunteer university students (15 men and 72 women; mean age + SE = 25.27 ± 2.99 years). Prior to enrolment, all participants were given a complete description of the study and signed a written informed consent. The study was approved by the Ethical Committee of the Department of Dynamic and Clinical Psychology, Sapienza, University of Rome (Prot. n. 0000453 and Prot. n. 0000112).

Clinical Assessment

Relationship Questionnaire (RQ). RQ (Bartholomew & Horowitz, 1991; Italian validated version by Scinto et al.,

1999) was used to measure attachment style. The RQ is a single-item measure made up of four short paragraphs, each describing a prototypical attachment pattern as it applies in close adult peer relationships. Participants are asked to rate their degree of correspondence to each prototype on a 7-point scale. The four attachment patterns (i.e. secure, preoccupied, fearful and dismissing) are defined in terms of two dimensions: anxiety (i.e. a strong need for care and attention from attachment figures coupled with a pervasive uncertainty about the willingness of attachment figures to respond to such needs) and avoidance (i.e. discomfort with psychological intimacy and the desire to maintain psychological independence). A cross-cultural study of the RQ conducted on a convenience sample of college students reported that the mean \pm s.d. score for the Italian population was 3.09 ± 2.01 (Schmitt et al., 2004). For our purpose we decided to use the RQ categorically, by dividing the four attachment styles in “secure attachment” ($N = 26$) and, on the other hand “insecure attachment” ($N = 61$), which includes fearful, preoccupied and dismissing attachment styles.

Difficulties in Emotion Regulation Scale (DERS). The DERS is a 36-item multidimensional self-report measure assessing individual’s characteristic patterns of emotion regulation. Items are rated on a 5-point Likert-type scale (from 1 = almost never to 5 = almost always) indicating the degree to which each statement describes the respondent’s behavior. Scores range from 36 to 180; greater scores on the DERS reflect greater difficulties with emotion regulation. This instrument consists of the following six subscales, theoretically formulated and confirmed through factor analysis: (1) Non-acceptance, referred to non-acceptance of emotion responses (e.g., “When I’m upset, I feel guilty for feeling that way”); (2) Goals, related to the difficulty in engaging in a goal-directed behavior while experiencing negative emotions (e.g., “When I’m upset, I have difficulty concentrating”); (3) Impulse, referring to the impulse control difficulty when experiencing negative emotions (e.g., “When I’m upset, I have difficulty controlling my behaviors”); (4) Awareness, related to emotional awareness (e.g., “I am attentive to my feelings”); (5) Strategies, concerning the limited access to emotion regulation strategies that are perceived as effective (e.g., “When I’m upset, I start to feel very bad about myself”); and (6) Clarity, related to the lack of emotional clarity (e.g., “I’m confused about how I feel”). The DERS showed a good level of internal consistency for both total score (Cronbach’s $\alpha = 0.93$) and the six subscales (Cronbach’s $\alpha > 0.80$) (Gratz & Roemer, 2004). The instrument also revealed an adequate concurrent validity with measures of emotion dysregulation and emotional avoidance, as well as a good predictive validity with behaviors associated with emotion dysregulation, such as self-harm and marital violence (Gratz & Roemer, 2004). For our purpose, the Italian validated version of the DERS was used (Girromini et al., 2012).

Biological sampling

Procedure for biological sampling. After the clinical assessment, saliva samples from voluntary university students were collected and transported to the laboratory for further processing.

Participants were asked not to drink, eat or smoke in the two hours prior to the saliva collection, in order to avoid/minimize sampling contamination.

DNA isolation and genotyping. Genomic DNA was extracted from saliva samples using a standard phenol/chloroform isolation procedure. DNA samples were genotyped for the A118G (rs1799971) SNP by the TaqMan® genotyping protocol. According to this protocol, 10 ng of DNA was poured into each well of the reaction plate, with 2.50 μ l of TaqMan® Universal PCR Master Mix (Catalog#: 4371353; Applied Biosystems, Branchburg, NJ) and 0.25 μ l of the corresponding TaqMan SNP genotyping assay (Catalog#: 4351379; Applied Biosystems), containing VIC and FAM probes. The plate was then run on an Applied Biosystems 7900HT Fast Real-Time PCR under the following thermal cycling conditions: an initial denaturation at 95°C for 10 min followed by 40 cycles of denaturation at 92°C for 15 s and annealing at 60°C for 60 s. Finally, genotypes were assigned by registering the fluorescence emissions from each well at the corresponding VIC and FAM dye wavelengths. Genotyping of each DNA sample was performed twice in blind conditions. For our purpose we decided to create two groups according to the presence of the homozygous A118G genotype variant A/A ($N = 63$), or the presence of the G-allele in the heterozygous A/G or homozygous G/G variant ($N = 24$).

Statistics

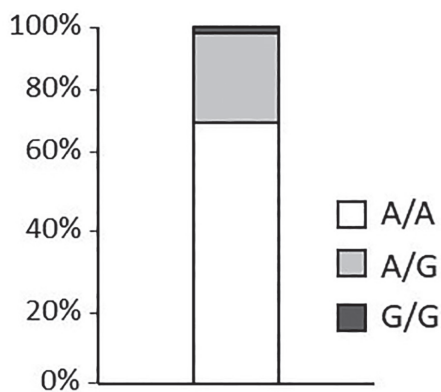
The effects of attachment style and A118G genotype on emotion regulation were first analyzed by multiple analysis of variance (MANOVA) followed by univariate ANOVAs and in cases of significance (<0.05), post-hoc comparisons using Tukey HSD’s test. Attachment styles and A118G genotype were used as categorical variables, while emotion regulation (DERS subscales) was used as continuous measure. Statistical analyses were carried out with the help of Statistica software Version 12.0 (StatSoft, Tulsa, OK, USA).

Results

A118G allele frequency distribution

The frequency distribution of the A118G genotypes in the sample was as follows: 73% A/A, 25% A/G, 2% G/G. Genotypic frequencies were in Hardy–Weinberg equilibrium (chi-square = 0.07, $df = 1$, $p = .79$). As already reported in other studies carried out on European/Caucasian populations (i.e., Sweeney et al., 2017; Troisi et al., 2012; The International HapMap 3 Consortium, 2010), the frequency of the G allele was very low with respect to the A one (approx. 15% vs. 85%). The G/G and A/G groups were therefore combined in the data analysis in a group of G-carriers (73% A and 27% G), according to the model of Arias, Feinn, and Kranzler (2006) and subsequent studies (Cimino et al. 2020; Copeland et al., 2011; Troisi et al., 2011, 2012. Figure 1).

Fig. 1. A118G allelic variants frequency distribution



Note. The overall distribution of the allelic variants is as follows: A/A 73%, A/G 25%, and G/G 2%.

A118G genotype modulates the impact of attachment style on emotion regulation

To determine if the A118G genotype modulates the impact of the attachment style on the emotion regulation a MANOVA of the DERS total score and subscales was performed. This analysis revealed a significant main effect of the attachment style ($\lambda = 0.76$; $F_{7,77} = 3.3$; $P = 0.002$). Univariate results showed a significant effect of the attachment style on DERS total score ($F[1, 86] = 17.189$, $P < 0.001$) and all DERS subscales: Non-acceptance ($F[1, 86] = 10.236$, $P = 0.002$), Goals ($F[1, 86] = 8.176$, $P = 0.005$), Impulse ($F[1, 86] = 11.023$, $P = 0.001$), Awareness ($F[1, 86] = 5.967$, $P = 0.016$), Strategies ($F[1, 86] = 20.526$, $P < 0.001$), and Clarity ($F[1, 86] = 15.707$, $P < 0.001$). As expected, insecure-attached individuals showed significantly higher scores in all these emotion regulation parameters than secure-attached individuals. A significant interaction effect between attachment style and A118G genotype on DERS

total score ($F[1, 86] = 4.018$, $P = 0.048$) and goals ($F[1, 86] = 4.279$, $P = 0.042$) was observed (Figure 2).

Specifically, the effect of attachment style was significantly more pronounced in G-allele carriers, with insecure-attached G-allele carriers showing significantly higher DERS total and Goals scores than secure-attached G-allele carriers. In all these subscales the G-allele carriers with insecure attachment showed the highest value among the four groups, whereas the G-allele carriers with secure attachment showed the lowest value among them (Figure 2; Table 1).

Tab. 1. Attachment categories and DERS total and goals scores

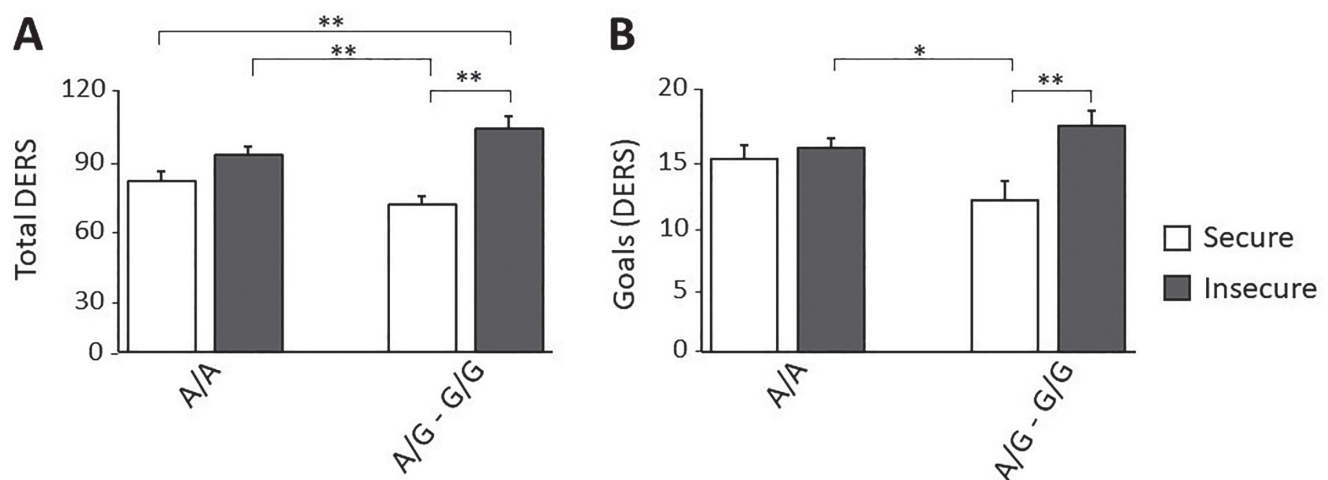
	Goals	Tot DERS
	Mean \pm St.Err	Mean \pm St.Err
A/A secure	14.60 \pm 1.09	78.26 \pm 4.21
A/A insecure	15.50 \pm 0.67	90.31 \pm 3.70
A/G-G/G secure	11.54 \pm 1.40	67.64 \pm 3.72
A/G-G/G insecure	17.15 \pm 1.37	102.23 \pm 5.62

Discussion

The results of this pilot study confirm that the MOR system, specifically the A118G gene, modulates the impact of the attachment style on the emotion regulation process.

Overall, a cross-cutting effect of the attachment on emotion regulation was noted. In particular, insecure-attached individuals exhibited the highest rates of difficulties in emotion regulation as assessed by the six DERS subscales (Gratz &

Fig. 2. A118G genotype modulates the impact of attachment style on emotion regulation



Note. ANOVAs revealed a significant interaction effect between A118G genotype and attachment style on the global score of the Difficulties in Emotion Regulation (DERS) (A) and Goals subscale (B). In particular, G-allele carriers with insecure attachment showed significantly higher DERS total and Goals scores than G-allele carriers with secure attachment. This effect was absent in the A-allele carriers. G-allele carriers with insecure attachment showed the highest value among the four groups, whereas the G-allele carriers with secure attachment showed the lowest value among them.

Roemer, 2004). These results are consistent with the large body of literature highlighting the association between attachment style and emotion regulation (Denham et al., 2010). Our data confirm that secure-attached individuals, unlike the insecure-attached individuals, exhibit a better management of negative emotions and they seem to activate more functional coping and emotion regulation strategies to alleviate distress (Kerns et al., 2000).

One limitation of this study is the low number of subjects that forced us to categorize the sample into secure vs insecure subjects thus preventing the investigation of the different patterns of emotion regulation manifested in the four different attachment styles. Previous reports have indeed shown that within the insecure attachment categories, anxious attached individuals tend to use hyperactivating strategies (defined by the amplification of proximity seeking strategies) to grab the attachment figure's attention and responsiveness (Winterheld, 2015; Brenning & Braet, 2013; Caldwell & Shaver, 2012; Burnette et al., 2009). On the contrary, avoidant-attached individuals apply deactivating strategies, by hiding the expressions of emotions and dealing with threats autonomously, with the aim of avoiding the frustration caused by significant other's emotional unavailability (Winterheld, 2015; Monti & Rudolph, 2014; Brenning & Braet, 2013; Caldwell & Shaver, 2012; Karreman & Vingerhoets, 2012; Simpson & Belsky, 2008).

This is the first study investigating the effect of A118G snp on emotion regulation and, although no effect of the A118G genotype was detected, an interesting interaction effect between attachment style and genotype was uncovered. The effect of attachment style was more pronounced in G-allele carriers, with insecure-attached G-allele carriers and secure-attached G-allele carriers showing, respectively, the highest and the lowest values in DERS total and Goals subscales among the four groups.

These results could be interpreted in light of the "Differential Susceptibility Model" formulated by Belsky and Pluess (2009). According to this framework, it seems that the G-allele acts as a plasticity factor to environmental and socio-relational stimuli. In our sample, the G allele seems to amplify the effects of the attachment style by producing emotion regulation difficulties in presence of adverse conditions, and, on the contrary, emotion regulation competence in presence of favorable conditions. This interpretation seems to be in line with previous evidence reporting that G-carriers exhibit increased dispositional sensitivity to social rejection (Way et al., 2009) and greater tendency to experience more pleasure from social and affective relationships with respect to A-carriers (Troisi et al., 2011). Further recent literature reports a higher frequency of insecure attachment in presence of low levels of maternal care, in G-carriers with respect to A-carriers (Cimino, 2020). In the context of adult attachment and romantic relationships, it has been reported that G-carriers show a stronger decrease in attachment security when interacting with a quarrelsome partner, compared to A-carriers (Tchalova et al., 2019). In biological terms this greater sensitivity to environmental (both positive and negative) stimulations of the G-carriers, with respect to A-carriers, appears to be linked to a lower brain MOR availability in this genotype (Krosiak et al., 2007)

Future studies would be therefore essential to clarify the relationship between MOR system and emotion regulation

also considering recent PET-imaging studies that document a correlation between MOR density, and their localization in specific brain structures, and emotional processing (Kantonen et al., 2020; Karjalainen et al., 2019). Finally, in light of the evidence that the two A118G variants show different sensitivity to the socio-relational stimuli, it would be interesting to assess how and if this genetic factor is able to modulate the psychotherapeutic relation, thus contributing to the clinical therapeutic success.

Compliance with Ethical Standards

Conflict of interest

The author declares that he/she has no competing interests.

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Ethical approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed Consent

Informed consent was obtained from all individual participants included in the study.

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