Social decision making, hierarchy and emotional contagion are interconnected by the basolateral amygdala

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Abstract

Decisions in social contexts might lead to choices favoring self- or others-interest, depending on the relationships between individuals. Prosocial and helping behaviors are evolutionary conserved across mammals. However, the neurobiological bases of choices that benefit others at a personal cost are not understood. Here, we revealed the role of the basolateral amygdala (BLA) in altruistic and selfish choices. We developed a two-choice social decision-making task in which mice could decide to share or not a positive reinforcement with their conspecifics. Preference for altruistic choices was more evident in males and if the conspecific was familiar. In particular, altruistic choices were associated with social dominance and affective state matching between individuals. Chemogenetic BLA neuronal silencing induced lower ranking hierarchy and less preference for altruistic choices. This provides a neurobiological comparative model of altruistic and selfish choices versus dominance hierarchy and emotional contagion, with relevance to pathologies associated with dysfunctions in social decision-making.

Introduction

Many of our most important decisions are made in the context of social interactions. These decisions require the integration of different cognitive processes and behaviors, which allow an individual to understand and interact with others[1,2]. The psychological conflict between self-interest and the interest of others, involving personal costs to benefit others, is a key element of social decisions that have an impact on daily life interindividual relationships[3]. From an evolutionary perspective, altruism likely evolved to promote survival through actions associated with kin selection, parental care, and reciprocity[4–6]. Although avoiding the term itself, increasing evidence from the fields of behavioral ecology, comparative psychology, and biology suggest that non-human animals engage in prosocial behaviors that resemble altruism. Parrots voluntarily cooperate for food rewards[7]; bonobos share food among companions[8–10]; rats help conspecifics that are in need[11–13], have been harmed[14,15], or are seeking food[16] and reciprocate previously received help[17,18]. Rodents display both consolatory[19] and collaborative[20] behaviors. Altruism is typically characterized by a direct personal cost to the altruistic individual with no conceivable long-term benefit. This capacity might differ from that to help or to prevent pain in others, captured by current animal paradigms[5,6,21,22], pointing to the need for exploration of whether animals choose altruism or self-interest, which could contribute to the understanding of the underlying neurobiology.

Mammals live in social groups with dominant and subordinate members, which determine a hierarchy that can affect multiple behaviors[23] and represent an important variable in social relationships and prosocial behaviors[24]. Moreover, socially close individuals share more easily subjective affective states of another trough emotional contagion[24,25]. All forms of empathy, including emotional contagion, are fundamental to adjusting one's own behavior with pro-social intention in group living animals[26]. Deficits in these processes are commonly observed in Alzheimer's Disease and frontotemporal dementia[27], psychiatric disorders[28] and psycho-pathological conditions[29], associated with dysfunctional social decision-making. Also cognitive factors might modulate decisions that impact on others involving

contextual information, such as past experiences and familiarity[21], and higher-level cognitive phenomena, such as perspective taking[30]. However, the neural bases of how these factors contributes to whether an individual choses altruism or self-interest remain poorly understood.

Research in decision making has growth in a variety of directions, identifying the neural bases involving both cortical and subcortical structures depending on different contexts and stimuli[31,32]. However, how social interaction and relationships might change decisions affecting selves and others among members of a group has been overlooked. Recent studies in non-human primates identified a prefrontal-amygdala network in which neural activity contribute to social decision-making[33]. Further, the same structures are involved in social interactions and social transmission in rodents[34,35]. In particular, the BLA has a central position in the neurobiological circuit for our abilities of choosing among options that differ in rewards and costs[36]. Yet, our understanding of its role in decisions involving altruism is still limited.

Here, we devised a social decision-making task (SDM) for mice that was modeled on the human game-theoretical paradigm known as the 'dictator game'[37] to specifically investigate decision-making in a social environment. Similar to the original paradigm, a 'dictator' (i.e., the actor) decides whether to share food with a 'recipient' (i.e., the observer), who is a passive player. We found that the majority of male mice, but not females, displayed a preference for sharing food with familiar, but not unfamiliar, conspecifics. Substantial individual differences in altruistic choices originated from the hierarchy status of each individual. Strikingly, chemogenetic silencing of the BLA abolished the development of altruistic choices. Furthermore, silencing of the BLA influenced hierarchical ranks within cagemates. Overall, we present a refinement to an experimental approach, which facilitated the exploration of various facets of altruistic-like behaviors in mice.

Results

Mice choose altruistic actions over selfish decisions

To test whether mice are capable of acting for the intentional benefit of conspecifics, we first devised a social decision-making task (SDM) for mice that was equivalent to the human "dictator game", one of the most prominent game-theoretical paradigms that have been designed to test altruism[37]. We expanded a standard operant cage with an adjacent compartment, separated by a metal mesh, in which to host a 'recipient' that would receive food rewards depending on the choice made by the 'dictator' (hereafter referred to as the 'actor'). The recipient was a passive player with a chance to receive a food reward from a magazine, depending on the actor's choice. To promote food-seeking behavior, at the start of the test, both the actors and the recipients were maintained at 90% of their free-feeding body weights. The actors were presented with a two-choice decision-making paradigm, in which nose poking resulted in either food rewards for themselves only (selfish choice) or for both themselves and the recipient (altruistic choice; **Fig. 1a**). We compared this condition against a control group of actor mice without the presence of a recipient. The structure of the task was identical between these two conditions ('with recipient' and 'no recipient'). Thus, any differences in the response could be attributed to the influence of the recipient.

Adult mouse littermates, three to six months-old, both males and females, were housed in same sex-pairs for at least two weeks before the start of testing. Animals were tested for five days, until they reached a stable performance for three consecutive days. At the group level, we found that actor mice with recipients preferred to share food rewards (altruistic choices) more frequently than not (selfish choices), exhibiting a positive decision preference index compared with that of mice in the 'no recipient 'condition, which did not display any choice preference (**Fig. 1b**). The location of the nose poke associated with altruistic or selfish response did not modified the preference for altruistic choices (**Fig. 1b**). Mice showed an increased number of altruistic over selfish responses when a recipient was present, whereas the mice in the 'no recipient' condition mice chose similarly between two nose pokes (**Fig. 1c**). Following the last session (day 5), we replaced the recipient mice with an inanimate object and tested the actors to determine whether any changes to their preference could be detected in the absence of social motivation. During this condition, the actors decreased their preference (both altruistic and selfish) in the presence of an inanimate object when compared against their behavior in the presence of the recipient (**Fig. 1d**). These results confirmed that the expression of the preference for altruistic or selfish choice was contingent on the presence of a conspecific.

We observed marked individual differences in the responses of the mice across days. We analyzed the performance of each actor separately and found that eleven of sixteen mice showed a significant increase in altruistic responses, more frequently than could be explained by chance (**Fig. 1e, f**), whereas the remaining five mice showed a significant decrease in altruistic responses (**Fig. 1e, f**). Altruistic and non-altruistic mice in the test condition that included a recipient showed significantly different choices starting on the second day of testing (Fig. 1d). The distribution of mouse preferences indicated that greater than 80% of the responses were altruistic for the majority of mice (**Fig. 1e**).

Next, we asked whether sex influenced altruistic behaviors during the SDM. We analyzed pairs of males and females separately. All actors, both males and females, displayed a significant preference (**Supplementary Fig. 1a**). At the group level, males displayed a significant preference for altruistic over selfish responses (**Supplementary Fig. 1a**) and only one male mouse of eight did not prefer to allocate food rewards to his recipient (**Supplementary Fig. 1b**). In contrast, the females did not show an overall preferential choice (**Supplementary Fig. 1a**). Among the eight tested pairs, half of the females displayed a preference for altruistic choices, whereas the other half made selfish choices (**Supplementary Fig. 1b**). Compared with the performance of sex-matched actors that performed the task in the absence of a recipient, only males showed a preference for the altruistic responses (**Supplementary Fig. 1c, d**).

To determine whether social interactions in the proximity of the divider between the actor and recipient compartments might have influenced the actors' decisions, we measured the time spent on social exploration in both mice and found that altruistic actor mice spent more time exploring their recipient than selfish actor mice (**Fig. 1g**). This was evident from the first session of testing, and this pattern was maintained until the last session (Day 5). In contrast, we did not observe any differences in the social exploration by the recipients (**Fig. 1h**). Importantly, we found that social exploration of the actor mice during the first day of testing was positively correlated with the altruistic responses on the last day of

testing, at which point the actors display a consistent behavioral preference (**Fig. 1h**). To test whether actor-recipient social interactions were necessary for the actors' social choices, we replaced the metal mesh with an opaque partition dividing the two compartments, which prevented social contact but allowed the passage of auditory and olfactory stimuli. We used a new cohort of mice and tested two conditions, one group of actor mice were tested with the opaque partition and the other group with the mesh. Mice tested in the presence of an opaque partition showed a significant decrease in altruistic responses compared with actor mice who were tested with a metal mesh that allowed social contact (**Fig. 1h**). These findings suggested that mice use social visual cues or social contacts to establish their decision preferences.

Finally, we tested whether sharing food with recipients could motivate a change in decision preference. The actors were first trained to trigger one of the two nose pokes, which both delivered the same food reward. After the mice displayed a stable preference for one nose poke, a recipient mouse was introduced to the adjacent compartment, and nose poking into the non-preferred hole resulted in the delivery of rewards to both mice, whereas nose poking into the preferred hole delivered rewards only to the actor (**Supplementary Fig. 2a**). The location of the recipient compartment did not biased actor mice preferences as nose poke responses during baseline training in the right and left nose-poke were not different (**Supplementary Fig. 2d**). At the group level, we found that actor mice displayed a positive change from their baseline preference across days, which suggested that the mice shifted their responses to share food rewards with their recipients (**Supplementary Fig. 2b**). Although we observed individual differences, the majority of mice displayed a switch from their preference ("altruistic," 8/13 mice, **Supplementary Fig. 2b, c**). Under this condition, the day following the last session, we replaced the recipient mouse with an inanimate object and found a decrease in preference compared with the preference expressed when the recipient was present (**Supplementary Fig 2f**). These results suggest that mice learned or were willing to change their behaviors to share a positive experience, such as a food reward, with their conspecifics.

Actor mice are willing to take altruistic actions even under costly conditions

To challenge the motivation of actor mice to allocate food rewards to their cage mates, we increased the cost of the altruistic decisions by reinforcing the responses at a fixed ratio of 2 (FR2, **Fig. 2a**). Under this condition, two nose pokes were required to receive food together with the recipient, whereas only one poke was necessary for selfish responses (FR1, **Fig. 2a**). We tested only those males and females mice that had previously demonstrated a significant preference above chance for altruistic responses after five days in the SDM (**Supplementary Fig. 1**). We similarly tested mice in the 'no recipient' condition, in which their natural preference was set to FR2, whereas the other nose poke option was maintained at FR1 (**Fig. 2a**).

Both males and females displayed an increased number of altruistic responses over selfish responses, even when additional effort was required (**Fig. 2b, c**). Moreover, male FR2 responses were higher than those performed by mice tested without a recipient (**Fig. 2d**). This difference was not confounded by the

baseline number of nose poke responses (**Fig. 2b**, *inset*). We then further increased the effort necessary to perform an altruistic action by increasing these responses to a FR4 (**Fig. 2a**). Under this condition, males showed increased altruistic responses compared to both females and to mice without recipients (**Fig. 2b, c**). Females did not show a preference between the two responses (**Fig. 2c**), and mice without a recipient switched their preference to nose poke reinforced at FR1 (**Fig. 2d**). When the altruistic responses were reinforced to FR6, the females switched their preference to the nose poke that delivered food rewards more easily (FR1, **Fig. 2d**), whereas males continued to prefer altruistic responses (**Fig. 2c**), making more responses using the FR6 nose poke compared with both female mice and mice without recipients (**Fig. 2b, c**). Finally, we tested males only in additional sessions to observe when a switch to the non-preferred response would occur. At FR8, male mice switched their preference to selfish responses, although they performed a similar number of nose pokes for both FR1 and FR8 (**Fig. 2d**). These results suggest that in the presence of a social motivation, male mice preferred to shared food rewards to benefit their cage mates, even under costly conditions.

In our setting, the altruistic responses were the results of conditioned learning, supported by a positive outcome for the actor. Thus, to dissect the social motivation to make an altruistic decision from the motivation to collect a food reward, we tested a satiety-induced reward devaluation. Mice were tested for five days in the SDM, and, following the last session, we devalued the reward outcome by pre-feeding the mice to satiety using the same reward pellets prior to test actor mice in a session that did not provide food rewards. We tested one condition in which neither the actor nor the recipient received rewards ('no reward'), whereas another group of actor mice was tested under conditions in which the actor mice did not receive any reinforcements but were still able to allocate food rewards to the recipient ('reward to recipient only'). Both groups of mice displayed reward devaluation, as indicated by a decrease in the nose poke response (**Fig 2e**), compared with previous session without pre-feeding (valued). However, we found that the mice increased their preference for altruistic responses when allocation to a recipient was possible, whereas mice that did not receive rewards and could not allocate rewards to the recipient did not modify their preference (**Fig 2e**). These results suggested that mice were willing to help their conspecifics, even in the absence of apparent self-interests associated with a positive reward outcome.

Mice display more selfish choices with unfamiliar conspecifics

Familiarity between individuals is known to amplify prosocial behaviors[11,19,38].To test whether social closeness affects the willingness to allocate food to others, in a new cohort of mice we tested the actions of actors in response to unfamiliar recipients that were housed in different cages. We found that actors tested in the presence of unfamiliar recipients showed opposite choices compared with actor mice tested in the presence of familiar recipients (**Fig. 3a**). In particular, actors (both males and females) made fewer altruistic responses in the presence of an unfamiliar compared to mice tested in the presence of cage mates (**Fig. 3b**). To determine individual differences in the responses across animals, we analyzed the performance of each actor. Under the condition featuring an unfamiliar recipient, we found that nine

mice of fifteen showed a significant increase in the number of selfish responses (**Fig. 3c, d**), whereas only three mice acted altruistically. Three mice did not show any preference (**Fig. 3c, d**). The distribution of mouse preferences showed that fewer than 20% of responses were altruistic for the majority of mice (**Fig. 3d**). Thus, when the actor mice were presented with non-cage mates, they acted more selfishly than actors paired with cage mates. These data indicated that familiarity facilitates altruistic choices in mice.

Social hierarchy differentiates preference for altruistic choices

Social animals self-organize into hierarchies, where group members vary in their level of dominance, affecting social relationship[24]. To determine the impact of the hierarchical relation between animals within the same cage on the preference for altruistic choices we used the tube test, a robust assay in which one mouse forces its opponent out of a narrow tube and classified dominant[39]. Mice were tested pairwise using a round robin design, on daily sessions after the SDM task, and the social rank of each mouse was calculated on the basis of winning against the other cage mates (**Fig. 4a**).

We analysed the relation of mice from 9 cages. In all cages the relation between mice was transitive and linear (a is more dominant over b, b more dominant g, g more dominant over d and then a should be dominant over all the others; **Fig. 4b**). Among the mice that performed the SDM task as actor and displayed preference for selfish choices, only 2 were a dominants, the rest were subordinate (**Fig. 4c**). Whereas, in the group of actor mice that displayed preference for altruistic choices, 6 were subordinate and 8 were dominant (**Fig. 4c**). We quantified this difference calculating a David's score (DS) for each mouse, a measure of dominance in the social hierarchy. Actor mice with preference for selfish choices displayed lower scores, and thus lower social rank, compared to their recipient conspecifics (**Fig. 4d**). Conversely, we did not detect any differences in DS between actor mice displaying preference for altruistic choices and their recipients (**Fig. 4e**). Finally, we grouped the actor mice that performed on the SDM task based on their social rank and found that mice in the a rank displayed higher altruistic preference compared to mice in the beta and gamma ranks (**Fig. 4f**). These results indicates that dominant individuals are more willing to share food rewards with their in-group members.

Emotional state matching motivates altruistic choices in dominant mice

Empathy refers to behavioral reactions to others emotional states, including the motivation to help and the affective tendency to experience the emotions of others[40]. Thus, we tested the hypothesis that the increased altruistic choices in familiar dominant mice could relate to an increased affective state matching between individuals. To do this we used an observational fear conditioning paradigm (**Fig. 4g**), where mice can vicariously match the emotional state of their companion[41]. Following the SDM task, actor mice and their recipients were placed in the two compartments of a double-chambered standard fear-conditioning apparatus, separated by a transparent partition. The actor mouse (observer) was allowed to observe the recipient (demonstrator) receiving repetitive foot shocks (**Fig. 4g**). We found that freezing behavior, which reflected the observational fear induced by social transmission, was higher in

altruistic versus selfish mice (**Fig. 4g**). Both groups of mice spent similar amount of time in exploration of their conspecific demonstrator (**Fig. 4g**). Furthermore, the scores obtained in the observational fear learning positively correlated with social dominance (**Fig. 4h**). Altogether these results indicate that dominant mice showed more empathic-like behaviors, including higher altruism and emotional contagion.

BLA neuronal silencing abolish emotional contagion

Encoding of information needed for social transfer have been reported to depend on neuronal projections from the anterior cingulate cortex to the BLA[34]. Thus, we first tested whether BLA downregulation could change the capacity to be affected by others affective state, using the observational fear conditioning paradigm (**Fig. 5a, b**). We used a chemogenetic approach to target the glutamatergic neurons in the mouse BLA with a virus carrying the inhibitory designer receptors exclusively activated by designer drugs (DREADD) receptor hM4Di (AAV-CaMKIIa-hM4Di-mCherry) an engineered inhibitory G-protein-coupled receptor that can decrease neuronal activity, or a control virus (AAV-CaMKIIa-mCherry, **Fig. 5a** and **Supplementary Fig. 3a, b**). To achieve neuronal downregulation in the BLA, hM4Di mice received clozapine-N-oxide (hM4Di CNO), 30 minutes before testing. As control, also the mice injected with AAV-CaMKIIa-mCherry received CNO (control CNO, **Fig. 5b**). We found a significant reduction of freezing behavior during the conditioning phase in animals treated with CNO for BLA downregulation, compared to control mice (**Fig. 5b**), suggesting reduced social transmission of emotions. Thus, consistent with a previous study[34], we show that the BLA is critically implicated in the emotional state matching.

BLA neuronal silencing revert the preference for altruistic choices

We revealed a correlation between emotional contagion and willingness to make altruistic choices. We then assessed whether BLA functioning can also mediate the latter process. Following the same approach used for the observational fear conditioning paradigm, we tested the effects of BLA neural silencing during the SDM task (**Fig. 5c**). We analysed the decision preference score of the actor mice of both control CNO and hM4Di CNO groups tested with familiar recipient mice. We found that whereas control mice displayed a positive score, suggesting preference for altruistic choices, BLA-silenced mice failed to show such preference (**Fig. 5d**). Indeed, control mice showed an increased number of altruistic over selfish responses, whereas mice with BLA-silencing did not show any difference between the two choices (**Fig. 5e**). Analysis of individual performance showed that six of the eight mice in which BLA was silenced displayed a significant preference for selfish choices (**Fig. 5f**). On the contrary, in control mice the majority (6/8) showed preference for altruistic choices (**Fig. 5f**), similarly to what we have shown in naïve animals tested with familiar recipients (**Fig. 1b**). The reduced preference for altruistic choices in BLA-downregulated mice was not dependent on reduced interest for social exploration (**Supplementary Fig. 3d, e**). Moreover, BLA silencing did not affect the number of responses and latency to make a choice (**Supplementary Fig. 3f, g**). Thus, these results indicate that the BLA is required to make decisions that benefit others. Altogether, these results indicate that BLA is crucially implicated in empathic-like behaviors, including increased altruism and emotional contagion.

BLA neuronal silencing reduces dominance

Social hierarchy influenced the preference for altruistic or selfish choices (**Fig. 4**). However, the implication of the BLA in the expression of social hierarchy is still unexplored. Thus, we tested whether the BLA could be linked also to the representation of social ranks. To test this, mice with CNO- induced BLA-silencing and control CNO mice were tested after the SDM in daily sessions in the tube test for the assessment of hierarchical relations (**Supplementary Fig. 4a, b**). All the recipient mice received as control AAV-CamKIIa-mCherry virus and were injected with CNO (**Supplementary Fig. 4a, b**). We found that silencing of the BLA in hM4Di CNO mice significantly decreased the dominance compared to control CNO mice (**Fig. 5g**). Indeed, a higher number of hM4Di CNO actor mice was subordinate to their recipient conspecific (**Fig. 5h** and **Supplementary Fig. 4c, d**). Further, whereas control mice were distributed across all the four ranks (a, b, g, d), mice with downregulated BLA ranked only b and g (**Supplementary Fig. 4e-g**). Altogether, consistent with our findings linking altruistic decision preference with hierarchy status, these experiments provide initial evidence of BLA as a common hub in the determination of social dominance, emotional contagion and social decision making.

Discussion

In this study, we showed that mice are willing to share food rewards with familiar conspecifics, even when these actions came at their cost or without any direct benefit. Divergence in social decision-making originated from differences in sex, emotional state matching and dominance hierarchy. In particular, we revealed in the BLA a shared neurobiological substrate needed to express emotional contagion, social dominance and preference for altruistic choices.

We developed an operant task modeled after a human game theory model, known as the dictator game, to explore how basic decision-making systems operate within a socially interactive environment. Most experimental studies of decision-making have examined behaviors with clearly defined probabilities and outcomes, such as choosing between food rewards. None of the previous studies in mice have added such complexity of social interactions[42]. However, most important decisions are made in the context of social interactions with others, dynamically changing based on feedbacks from others. Thus, this social decision-making paradigm may offer a complementary approach for the examination of distinct and more complex social behaviors. In our task, mice chose between two actions, either yielding a reward only for themselves, or yielding a reward both for themselves and for a partner placed in an adjacent compartment. The majority of mice developed a clear preference for one of the two choices. At the population level, actor mice paired with familiar conspecifics preferred the altruistic action, whereas they chose the more selfish action when offered the opportunity to share food rewards with unfamiliar others. Altruistic behaviors are thought to have evolved to help family members or those inclined to return the favor[43]. Thus, familiarity bias further suggested that the choice to share food with others was intentional, as there would be no reason to prefer altruistic choices with familiar compared to unfamiliar conspecifics.

We confirmed that preference for altruistic choices was contingent on the presence of a conspecific, by testing actor mice with inanimate objects, without recipients, and with hidden recipients. Our results are consistent with recent studies performed in rodents, which demonstrated complex pro-social behaviors in rats, such as preference for mutual rewards[44], helping behaviors[11] and the avoidance of harming others[14]. However, fewer studies have demonstrated that mice are capable of displaying pro-social behaviors reminiscent of altruism constructs. Rodents spontaneously console or help distressed conspecifics[19,45], and have been shown to intentionally cooperate for the reciprocal exchange of services[20], which suggested that they can act to increase mutual benefits. Compared with these studies, in our task, mice learned to make altruistic choices through conditioned responses that were reinforced by a positive outcome also for the actor. Nevertheless, the mice displayed an interest in sharing food with their conspecifics even in the absence of any explicit return favors from their actions, which suggested that the pro-social choices persisted without evident self-benefit that is a critical factor that defines "altruistic" choices[46]. Moreover, when mice were presented with the opportunity to stop making altruistic choices by exerting a less effort to obtain a food reward only for themselves, they continued to display a preference for sharing food with their companions, even when this action was associated with unfavorable conditions. Altogether these findings might suggest that mice perceive their action as rewarding because of the effect on their conspecifics.

Our behavioral results showed that dominance hierarchy contributed to the preference for altruistic of selfish choice. Social status is very important in a social group and can guide behavior and motivation, including in humans[47]. Here, we found that the majority of mice that displayed preference for selfish over altruistic choices were subordinate to their recipient. This could be interpreted as a competition for food, as dominant members might benefit from easier access to food[48]. In non-human primates, where social ranks are very strict and stable, prosocial responses were more often directed from dominant towards subordinate members[24]. Thus, dominant individuals would behave in ways that benefit others in order to advertise their dominance. Although many species differences may exist[24], this is in line with our finding that mice ranked a were on average more altruistic. Interestingly, mice that displayed more selfish choices ranked on b and g, whereas a and d displayed more altruistic preferences. This may indicate that mice on the intermediate status (b, g) are more selfish because in competition for both upward and downward ranks, compared to lowest status members d. Thus, our task generated a variety of behavioral responses that could address a number of complex aspects of social decision-making, triggered by interpersonal interactions and potentially associated with emotions, personality, and motivation.

The role of BLA in decision-making has been largely investigated in rodents under several conditions, such as risk-taking, punishments and threats[36,49]. This evidence provides a robust picture of the critical role of the BLA in the integration of reward-related information and costs to guide decision-making. Association of these information are also integrated by BLA with motivational and emotional inputs from prefrontal and insular cortices[50]. In non-human primates synchronization of neural activity between BLA and prefrontal cortex is important for the establishment of other-regarding preference[33]. Thus, we could hypothesize that perturbation of BLA neuronal activity during social-decision making might had affected

a similar pathway in mice causing desynchronization of these brain regions. The preferential connection between BLA and cortical structures, such as the prefrontal cortex, has an important modulatory effect also on social behavior and transmission of social cues[34,35]. In agreement, we found that downregulation of BLA reduce the capacity to be affected by emotional states of others, which was correlated with preferences on social decision-making task. Thus, the establishment of a preference toward altruistic or selfish choices could be at least in part be related to empathy-like capacity in mice. Altogether, the effects observed following neuronal silencing of the BLA might indicate that the BLA could mirror the value of reward for self and for others.

Downregulation of the BLA was also associated with lower social dominance scores. Our understanding of the neural mechanisms associated with social hierarchy is limited, although, previous studies provided evidence pointing to the involvement of the prefrontal cortex[51,52]. Furthermore, in non-human primates neural ensembles in the amygdala are correlated with the social rank of conspecific images[53] and ablation of the amygdala caused a change from top to bottom of the dominance hierarchy[54]. In this study we help to complete previous findings by indicating that the BLA is involved in the representation of social status that contributed to the expression of preferences in the social decision-making. Indeed, animals with silencing of the BLA displayed higher preference for selfish choices, consistent with our results in naïve mice showing that subordinate actor mice express more likely preference for selfish choices. Interestingly, animals treated with CNO for BLA downregulation all ranked b and g, which we also showed to be among the animals that express lower preference for altruistic choices. Taken together, these data indicate that the BLA carry information about social rank that is critical for social decision-making. Considering the involvement of the PFC in the plastic modulation of social hierarchy[51], and the role of the ACC in social information processing and empathy[55], and the reciprocal connections between the BLA and these cortical regions, the BLA could be an hub of a network where information are gathered to guide prosocial behavior.

Altruistic behaviors were learned in our task through positive reinforcements, nevertheless we cannot rule out the possibility that some degree of innate or impulsive altruism may have assisted during the initial learning process[21]. In support of this possibility, we found that mice that expressed a preference for altruistic choices were more interested in social exploration than mice that preferred selfish choices. The motivation to act in an altruistic manner, from a biological point of view, has been suggested to be explained by kin selection, in which the advantage occurring to close kin would be what makes the altruistic behavior selectively advantageous[21]. However, in a laboratory setting, animals do not face such selection pressures, although the animals may experience mild food restrictions; therefore, other explanations should be considered, such as, empathy-related behaviors, including emotional engagement between the actor and recipient. Mice are able to sense[41,56] and discriminate[25] the affective state of their conspecifics. In agreement, we found that mice that expressed preference for altruistic choices displayed more empathy-like behaviors. Food seeking behavior of the recipient could trigger an emotional transfer between mice, which may motivate altruism. Moreover, familiarity can amplify the empathic response[26].

Consistently, we found that emotional contagion was linked with the preference for altruistic choices were positively correlated, suggesting that in mice affective state matching could drive altruism.

Together with empathy, social motivation could represent another explanation for altruism. Rodents are social animals[57] that express preferences for social closeness and avoid social isolation[58], which can have rewarding properties[59]. Moreover, positive experiences with other animal have been shown to augment altruistic behaviors[26]. For example, rats faced with the dilemma of whether to free a constrained conspecific or to gain access to chocolate were more likely to free their distressed conspecifics and share the chocolate[11]. This interpretation is consistent with the observation that mice displayed a willingness to help their companions under unfavorable conditions, such as exerting the increased effort necessary to share food rewards, even when the same reward could be obtained with less effort. This result suggested that an additional underlying motivation may exist for this behavior, which could include the well-being conferred by sharing a positive experience, similar to that experienced when helping others.

In summary, we developed a task enabling to detect in mice individual differences in the preference for sharing positive reinforcements with others. Using this task we investigated the neurobiology of social decision making across aspects associated with altruistic and selfish choices, reveling that the BLA carry information about social dominance and emotional contagion which are all critical for social decision-making. These results could have important implications for psychiatric, psychological, and neurodevelopmental conditions associated with disruptions in social decision-making.

Declarations

Author contributions

Conceptualization: DS. Methodology and Investigation: DS, FLG, FM. Resource: DS, MD, FP. Writing: DS, FLG, MD, FP. Visualization and Analysis: DS, FLG, FM, NC. Supervision: DS, MD, FP. All the authors revised the manuscript.

Competing interests

The authors declare that they have no competing interests.

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Methods

Mice. All procedures were approved by the Italian Ministry of Health (permits n. 107/2015-PR and 749/2017-PR and 191/2020-PR) and local Animal Use Committee and were conducted in accordance with the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health and the European Community Council Directives. Routine veterinary care and animals' maintenance was provided by dedicated and trained personnel. Three to five-month-old males and females C57BL/6J animals were used. Distinct cohorts of naïve mice were used for each experiment. Animals were housed two to four per cage in a climate-controlled facility (22±2 C), with *ad libitum* access to food and water throughout, and with a 12-hour light/dark cycle (7pm/7am schedule). Experiments were run during the light phase (within 10am-5pm). All mice were handled on alternate days during the week preceding the first behavioral testing.

Behavioral paradigm.

Social decision-making task. <u>Experimental setup.</u> Experiments were conducted in a standard operant chamber (actor's compartment, L: 24 cm x W: 20 cm x H: 18,5 cm; ENV-307W-CT; Med Associates, Inc.) fused with a custom-made small triangle-shaped chamber that hosted the recipient (L: 18 x W: 14 cm x H: 18,5). The separation wall between the compartments (operant chamber and recipient chamber) was replaced by a metal mesh with 1cm holes that allowed social exploration and nose-to-nose interaction. The actor's compartment was equipped with two nose poke holes and a food magazine between them, for delivery of food rewards (14 mg; Test Diet, 5-TUL). The recipient's compartment presented only a food magazine connected to a food dispenser. The setup was placed inside a sound attenuating cubicle (ENV-022V, Med Associates, Inc) homogeneously and dimly lit (6 ± 1 lux) to minimize gradients in light,

temperature, sound and other environmental conditions that could produce a side preference. All tasks were controlled by custom scripts written in MED-PC IV (Med Associates, Inc.). A digital camera (Imaging Source, DMK 22AUC03 monochrome) was placed on top of the setup to record the test using a behavioral tracking system (Anymaze 6.0, Stoelting).

Task design. The testing subjects, the "actors", were tested in three different conditions: *i*) with recipient, in which a cage mate was placed in the adjacent compartment, and acted as recipient; *ii*) no recipient, the compartment of the recipient was empty; *iii*) with toy, the recipient was replaced with an inanimate object. The actor (A, Figure 1a) determined to receive a food reward for himself (selfish choice) or to allocate the reward also to his companion (altruistic choice), "the recipient" (R). Both choices were reinforced on fixed ratio 1, such that poking into the left or right nose poke resulted in one food reward delivery. Altruistic and selfish responses were counterbalanced between left and right nose pokes across mice. After one nose poke, an intertrial interval of 5 seconds occurred. The recipient was a passive player and only received food rewards upon actor choices. In the no recipient condition, the adjacent compartment was empty, while in the toy condition, an inanimate black object was placed in the recipient compartment. The task design was identical across the condition with recipient and served as a control for pellet delivery sounds and for potential secondary effects of reinforcement.

Actor and recipient were mildly food-restricted to 90% of their baseline weight and were housed together for at least two weeks before the experiment. In the condition with unfamiliar recipient, actor and recipient were never housed together. The actors were tested for five days, in 40 minutes sessions, with a partner (with recipient) or without (no recipient), with an inanimate object, or with an opaque partition dividing recipient and actor, depending on the testing condition. Actors were always paired with the same recipient throughout the same experiment. In the toy condition, actors were tested for five days with a partner and the day following the last session (day 5), the recipient was replaced by an inanimate object (day 6). In condition with the opaque partition between actor and recipient actor mice were tested for five days. The opaque partition did not allow visual cues and social exploration/interaction.

Fixed ratio schedules. To test whether mice made voluntary choices to benefit others under costly conditions, we tested mice using increasing fixed ratio (FR) schedule for altruistic decisions from FR2 to FR8. In this condition, the number of operant responses required to dispense food to the recipient is increased on each day (from 2 to 8). Selfish responses remained on FR1 throughout the experiment. In the 'no recipient' condition, for each actor the preferred nose poke was reinforced using the increasing FR schedule and the other nose poke was kept on FR1.

Satiety-induced reward devaluation. Mice were tested for five days in the SDM and following the last session, actors were singly housed for one hour and reward outcome was devalued by pre-feeding them to satiety giving free access to reward pellets in their cage. Then, mice were transferred to the operant chamber and test in a non-reinforced session. Two groups of mice were tested: in one condition, both actor and recipient did not receive rewards ('no reward') and in the other, actor mice did not receive any reinforcement, but they could still allocate food rewards to their recipients ('reward to recipient only').

Analyses. The number of nose poke responses was counted by a software (MED-PC V, Med Associates, Inc). To quantify individual preferences of altruistic over selfish responses or left and right nose pokes we calculated a decision preference score, as following: (number of altruistic responses - number of selfish responses) / total number of responses. Video images were analyzed *a posteriori* for scoring of exploratory behavior using Anymaze 6.2 (Stoelting, UK) and Boris[60]. We measured the time spent by the recipient and the actor in social exploration in the area in the proximity of to the adjacent compartment where they could explore each other.

Tube test. The tube test was performed as described in a previous study[61]. We used a transparent Plexiglas tube (L: 30 cm, inside diameter 3 cm). For habituation, the tube was placed inside the cage for three consecutive days. After habituation, mice were trained to run inside the tube. Each mouse was released at alternating ends of the tube and was allowed to run through the tube. We used a plastic stick to guide the mouse to the end of the tube if needed. Each animal was given ten training trials on two consecutive days. For the test, two mice were simultaneously released into the opposite ends of the tube and care was taken to ensure that they met in the middle of the tube. The first mouse that retreated and placed its two rear paws outside the tube was recorded as the "loser" of the trial and the other mouse the "winner". Between each trial, tube was cleaned with 75% ethanol. Mice were tested pairwise using a round robin tournament, on daily sessions. Each pair of cage-mates was tested in consecutive trials, alternating the starting side of the tube. The test was performed until all the ranks were stable for at least 4 continuous daily trials. To assign each animal social rank we used the normalized David's score (DS) for dominance. The score was calculated from the individual proportion of wins and losses in all the trials, in relation to the wins and losses of its opponents, as reported in a previous study[62]. We then normalized the score to be between 0 and *N-1* (where *N* is the number of subjects in each cage), using the following formula:

Observational fear conditioning. The apparatus consisted of two identical and adjacent fear conditioning chambers (Ugo Basile, 24×20×30 cm) separated by a transparent Plexiglas partition. Olfactory and auditory cues could be transmitted between the chambers. A demonstrator mouse (previously recipient in the SDM task) and an observer (previously actor in the SDM task) were individually placed in the two chambers and allowed to explore the chambers for 5 min (baseline). Then, a 2-s foot shock (0.7 mA) was delivered every 10 s for 4 min to the demonstrator mouse using a behavior tracking software (Anymaze 6.0, Stoelting). The same pairs tested in the SDM were used. Based on previous studies[28], we used 10-s intervals for foot shocks and a 4-min training. At the end of the procedure mice returned to their home-cage.

Viral injections. *Viral vectors.* AAV5-CamKIIa-mCherry (114469, titer ≥ 7×10¹² vg/mL) and AAV5-CamKIIa-hM4D(Gi)-mCherry (50477, titer ≥ 3×10¹² vg/mL) were purchased from Addgene.

Surgical procedures. C57BL/6J mice were naïve and 2 months old at the time of surgery. All mice were anesthetized with a mix of isoflurane/oxygen 2%/1.5% by inhalation and mounted into a stereotaxic frame (Stoelting) linked to a digital micromanipulator. Brain coordinates of viral injection in the BLA were

chosen in accordance with the mouse brain atlas[63]: AP: -1.7 mm; ML: ± 3 mm; DV: -4.5 mm. The volume of AAV injection was 0.4 mL per hemisphere. We infused virus through a 10-µL Hamilton syringe. After infusion, the pipette was kept in place for 5 min. After virus injection mice were allowed 4 weeks to recover and for the viral transgenes to adequately express before behavioral experiments.

Drugs

For hM4D activation, to achieve BLA downregulation we used i.p. administration of Clozapine N-Oxide dihydrochloride (CNO, HB6149 Hello Bio) dissolved in physiological saline (0.9% NaCl) at a dose of 3mg/kg in a volume of 10 ml/kg, 30 minutes before the behavioral experiments. All mice (control CNO, hM4D CNO) received i.p. CNO injection.

Histology

To assess localization and expression of viruses, mice were perfused with 40 ml of PBS and then 40 ml of PFA 4% in PBS. Brains were then postfixed in PFA 4% in PBS for 1 hour at 4°C and cut into 50 mm-thick slices with a vibratome. BLA-containing brain slices were acquired with Nanozoomer S60 (Hamamatsu) to detect native fluorescence of the mCherry-fused chemoreceptor, using constant settings.

Statistics. Results are expressed as mean ± standard error of the mean (s.e.m.) throughout the manuscript. For the analysis of decision preference score and percentage of altruistic responses we used two-way repeated measures ANOVA, followed by Bonferroni multiple comparison test. For the analysis of number of nose poke responses, we used two-way ANOVA, followed by Bonferroni multiple comparison test. For the analysis of social exploration, we used two-way repeated measures ANOVA, followed by Bonferroni multiple comparison test. Mice were assigned to altruistic or selfish groups using one sample t-test to chance (50%). The accepted value for significance was $p<0.05$. Statistical analyses were performed using GraphPad Prism 9. Numbers of mice are reported in the figure legends. Data distribution was tested using D'Agostino and Pearson normality test. The experiments reported in this work were repeated independently two to four time. No statistical methods were used to predetermine sample size for single experiments. The animal number were based on estimation from previous studies. Littermates were randomly assigned to the different groups. Experimenters were not blinded during data acquisition, but all analyses were performed with blinding of the experimental conditions as stated in the methods section.

Data availability. The data that support the findings of this study are available from the corresponding author upon reasonable request.
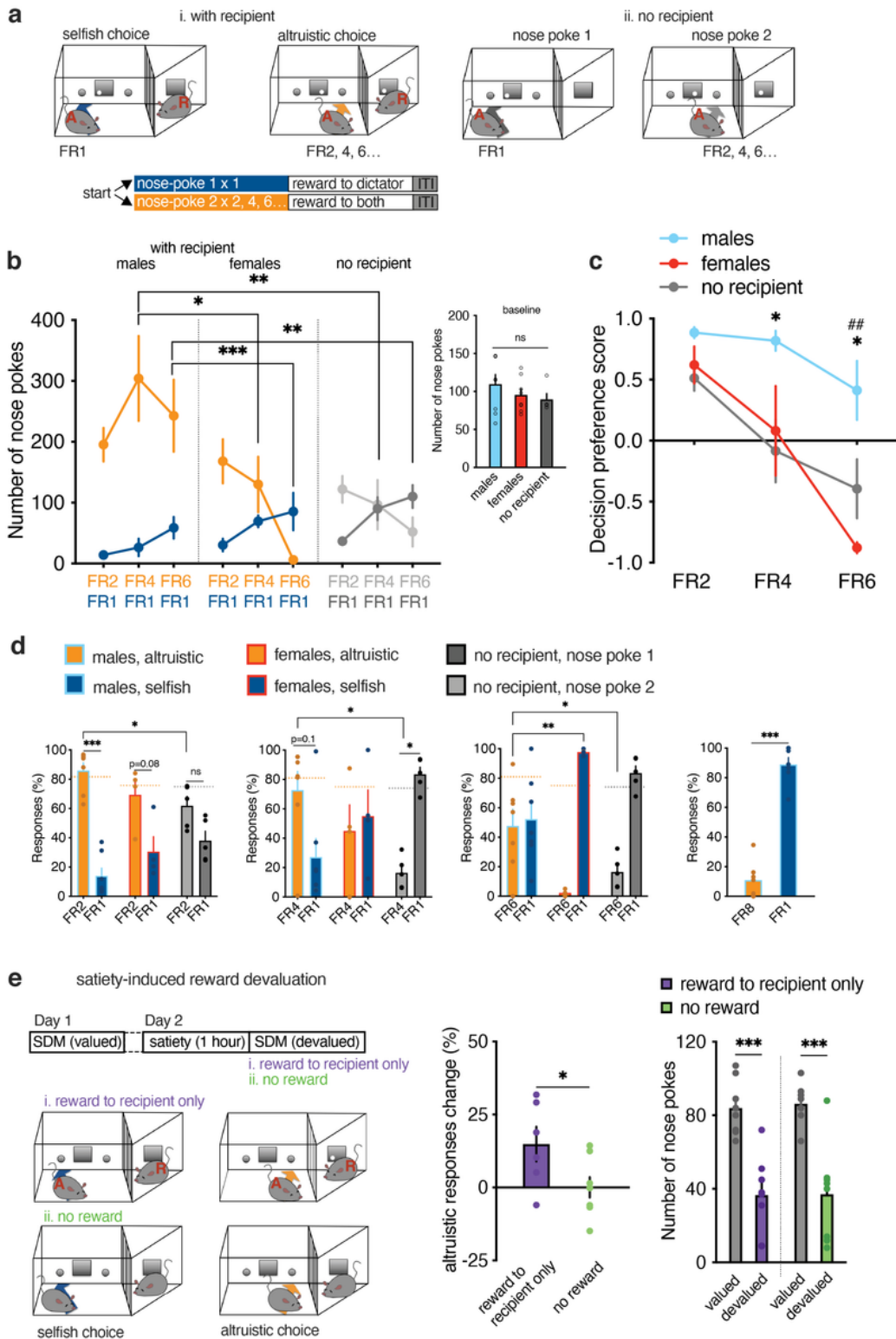
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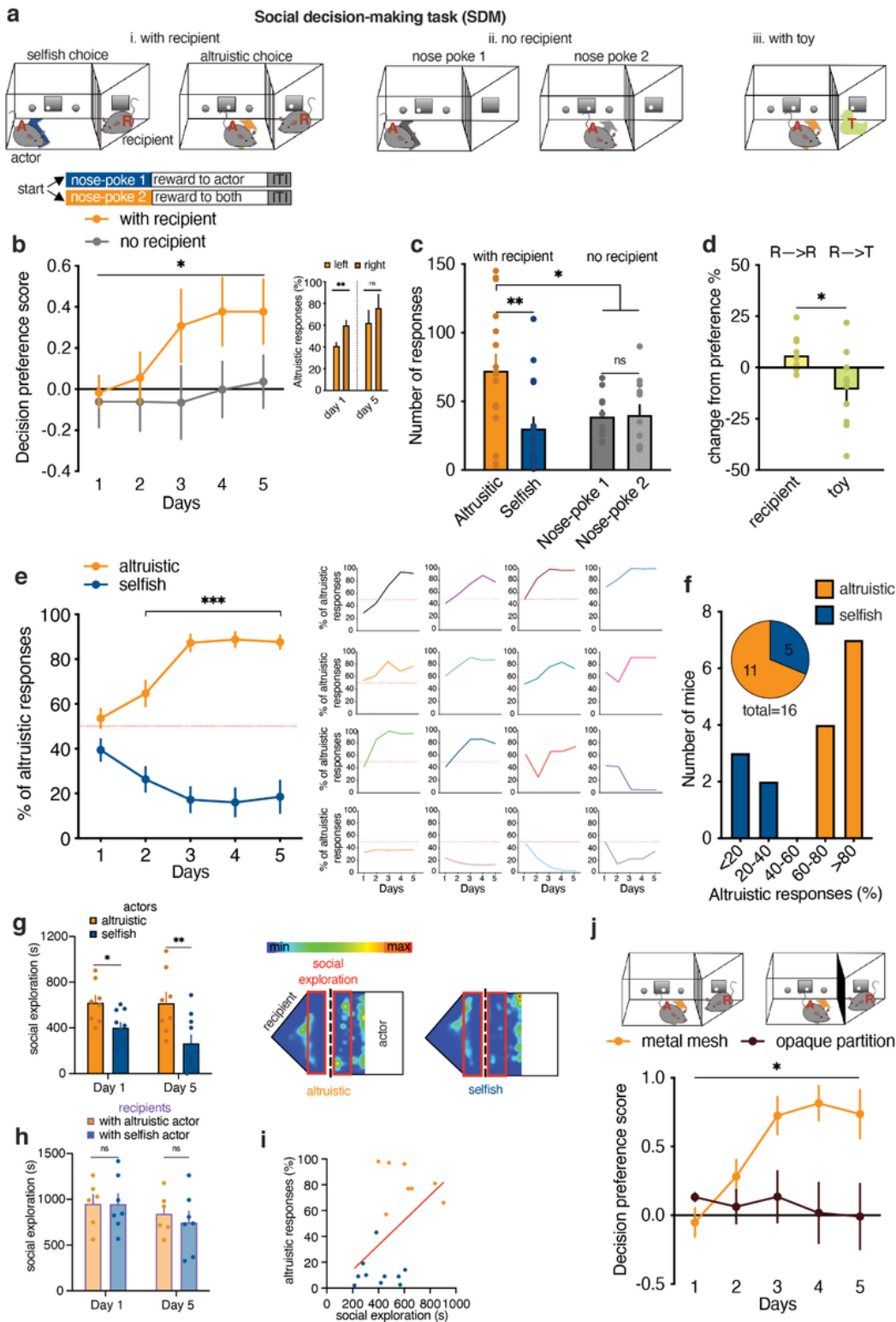
Figure 1

Mice prefer altruistic over selfish decisions. a, experimental design of the SDM. Actor mice were trained on a two-choice decision paradigm where nose pokes resulted in food rewards. In the condition i.'with recipient' (orange) one nose poke resulted in food reward to actor (selfish choice) and the other nose poke in food reward both to the actor and to the recipient, in the adjacent compartment (altruistic choice). After an inter-trial interval of 5 seconds (ITI), a new trial started, and actor could make their choice. The location

of the two responses were counterbalanced between left and right nose-pokes. In the condition ii. 'with recipient' (grey) the structure of the task was identical, but the adjacent compartment was empty. iii. In the condition 'with toy', the recipient was replaced with an inanimate object. b, decision preference score in the five days of SDM in mice tested with recipient or no recipient (two-way RM ANOVA, group (with recipient, no recipient) x time (days 1-5), $F_{(4, 104)}=2.485$, $p=0.0481$; the decision preference scores, for both conditions, were found to fit a normal distribution across five days of testing, D'Agostino and Pearson normality test, 'with recipient', min $K2=2.98$, $p=0.225$; 'no recipient' min $K2=0.87$, $p=0.071$; n=16, males/females 8/8). Inset, altruistic responses on day 1 grouped by choices on left and right (two-tailed unpaired t-test, $t=3.37$, d.f.=14, $p=0.0046$) and on day 5 ($t=0.79$, d.f.=14, $p=0.4419$). c, number of nose poke responses in the condition with recipient (altruistic, orange and selfish, blue) and no recipient (nose poke 1, dark grey and nose poke 2, light grey); two-way RM ANOVA, group (with recipient, no recipient) x response (nose poke 1, nose poke 2), $F_{(1, 52)}=5.766$, $p=0.019$). d, change of preference when animals were tested one additional day with their recipient (R R) or with an inanimate object (toy, R T) (two-tailed unpaired t-test, $t=2.24$, d.f.=18, $p=0.0374$, n=10 each group). e, mice were assigned to altruistic (orange) or selfish (blue) using one sample t-test to chance (50%, red line). Altruistic responses (in %) in altruistic and selfish mice (two-way RM ANOVA, group (altruistic, selfish) x time (days 1-5), $F_{(4, 56)}=21.55$, $p<0.0001$) and individual scores of altruistic responses across five days of SDM. f, number of tested mice grouped by percentage of altruistic responses. g, (Left) social exploration (in seconds) of altruistic (orange) and selfish (blue) actors towards their recipients during first (Day 1) and last (Day 5) session of SDM (two-way ANOVA, group (altruistic, selfish), $F_{(1, 32)}=16.29$, $p=0.0003$; n=8/10 each group). (Right) Schematic illustration of the actor-recipient testing chambers with graphical representation of the amount of time actors spent in different parts of the chamber (with blue as the shortest and red as the longest time). Social exploration was measured in the area highlighted in red. h, social exploration of recipients towards altruistic (orange) or selfish (blue) actors during first (Day 1) and last (Day 5) session of SDM (two-way ANOVA, group (altruistic, selfish), $F_{(1, 11)}=0.16$, $p=0.6902$; n=6/7 each group). i, correlation between social exploration on day 1 and preference for altruistic decisions (percentage of altruistic responses, day 5, $r=0.4890$, $p=0.039$, n=18 pairs). j, altruistic responses (%) in mice separated from the recipients by a metal mesh (orange) or an opaque partition during (maroon) the SDM (two-way RM ANOVA, group (metal mesh, opaque partition) x time (days 1-5), $F_{(4, 56)}=3.8$, $p=0.008$, n=6/10 each group). *$p<0.05$, **$p<0.01$, ***$p<0.001$. n.s. not significant. Values are expressed as mean ± s.e.m.

Figure 2

Mice are willing to take altruistic decisions under costly situations. a, altruistic responses were reinforced on a fixed ratio (FR) schedule that increased by two responses across days, starting from FR2. Selfish responses were reinforced on a fixed ratio 1 (FR1) for the entire duration of the test. We tested only mice that showed significant preference above change for altruistic responses after five days in the SDM; (n=11, males/females 7/4. Mice in the 'no recipient' condition (n=6) were tested on the same FR schedule

on the preferred nose poke after 5 days of SDM without recipient (males and females were pooled together as we did not find statistical difference). b, Left, Number of nose poke responses on FR1 versus FR2, FR4 and FR6 in males and females actors and actor tested without recipient (between groups: two-way RM ANOVA, group (with recipient males, with recipient females, no recipient) x response (FR2, FR4, FR6), $F_{(10, 52)}=4.25$, $p=0.0002$; within groups: two-way RM ANOVA, group (with recipient males, with recipient females, no recipient) x response (FR2, FR4, FR6), $F_{(4, 26)}=4.48$, $p=0.0069$). Right, number of nose pokes on day 5 of the SDM (one-way ANOVA, $F_{(2, 18)}=0.91$, $p=0.4192$). c, decision preference score of responses on FR2, FR4 and FR6, compared to FR1, in mice tested with the recipient (male, light blue, and female, red) and mice in the no recipient condition (grey) (two-way ANOVA, group (with recipient males, with recipient females, no recipient) x response (FR2, FR4, FR6), $F_{(4, 26)}=3.55$, $p=0.0193$). males: *$p<0.05$ vs. no recipient, ##$p<0.005$ vs females. d, altruistic responses (orange) reinforced on FR2, FR4 and FR6 and selfish responses (blue) reinforced on FR1 expressed as percentage of the total in males (light blue) and females (red) mice and responses on the preferred nose poke (NP1, dark grey) reinforced on FR2, FR4 and FR6 and responses on the non-preferred nose poke (NP2, light grey) reinforced on FR1 in mice tested without recipient (FR2: two-way RM ANOVA, group (with recipient males, with recipient females, no recipient) x response (FR1, FR2), $F_{(2, 13)}=3.5$, $p=0.05$. FR4: two-way RM ANOVA, group (with recipient males, with recipient females, no recipient) x response (FR1, FR2), $F_{(2, 13)}=5.1$, $p=0.0192$; FR6. two-way RM ANOVA, group (with recipient males, with recipient females, no recipient) x response (FR1, FR2), $F_{(2, 13)}=6.6$, $p=0.0103$. FR8: two-tailed unpaired t-test, $t=8.32$, d.f.=6, $p=0.0002$). e, satiety-induced reward devaluation. Following the last session of SDM (day 5, "valued" session) reward outcome was devalued by pre-feeding mice to satiety with the same reward pellets and then test on a non-reinforced session. Change in altruistic choices (%) during devaluation test in actors that could allocate rewards to recipients (but not for them-selves, magenta) and in mice that did not receive any rewards (green, two-tailed paired t-test: $t=2.28$, d.f.=12, $p=0.0410$), and number of nose pokes responses during valued and devalued sessions (two-way RM ANOVA, session type (valued, devalued), $F_{(1, 14)}=43.07$, $p<0.0001$). *$p<0.05$, **$p<0.01$, ***$p<0.001$. n.s. not significant. Values are expressed as mean ± s.e.m.
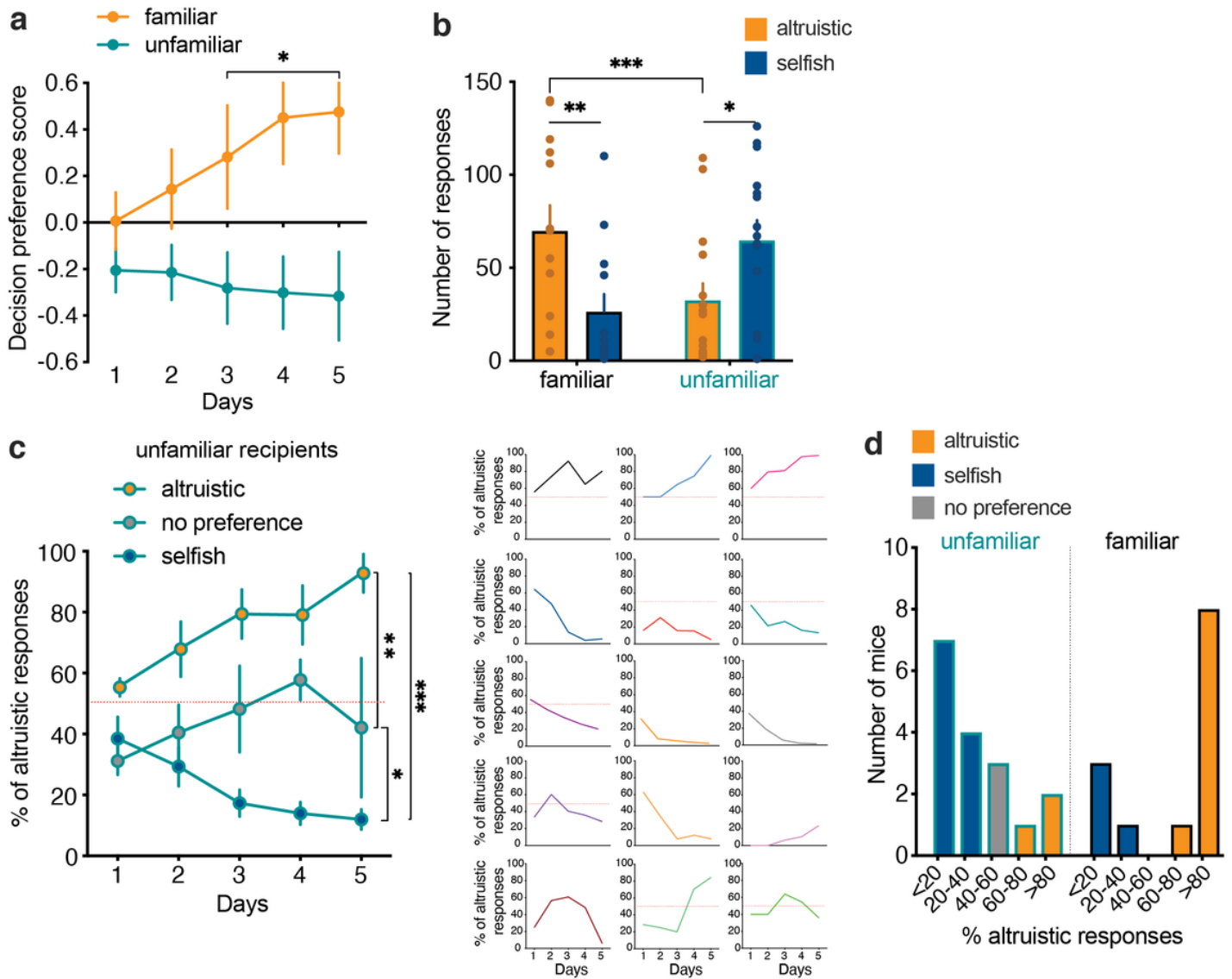
Figure 3

Mice display more selfish choices with unfamiliar conspecifics. a, decision preference score in the five days of SDM in mice (n=15, males/females 10/5) tested with familiar (orange) or unfamiliar (green) recipients (two-way RM ANOVA, group (familiar recipient, unfamiliar recipient) x time (days 1-5), $F_{(4, 104)}$=2.707, p=0.0342). b, number of nose poke responses in the condition with recipient (altruistic, orange and selfish, blue) and no recipient (nose poke 1, dark grey and nose poke 2, light grey; two-way RM ANOVA, group (familiar, unfamiliar) x response (altruistic, selfish), $F_{(1, 52)}$=12.03, p=0.0011). c, mice were assigned to altruistic (orange), selfish (blue) or no preference (gray) using one sample t-test to chance (50%, red line). Altruistic responses (in %) in altruistic, selfish and mice that did not display a preference (two-way RM ANOVA, group (altruistic, selfish, no preference) x time (days 1-5), $F_{(8, 48)}$=4.22, p=0.0007) and individual scores of altruistic responses across five days of SDM. d, number of mice tested with an unfamiliar recipient grouped by percentage of altruistic responses (chi-square test, $\chi 2$=5.99, p=0.0143). *p<0.05, **p<0.01, ***p<0.001. n.s. not significant. Values are expressed as mean ± s.e.m.
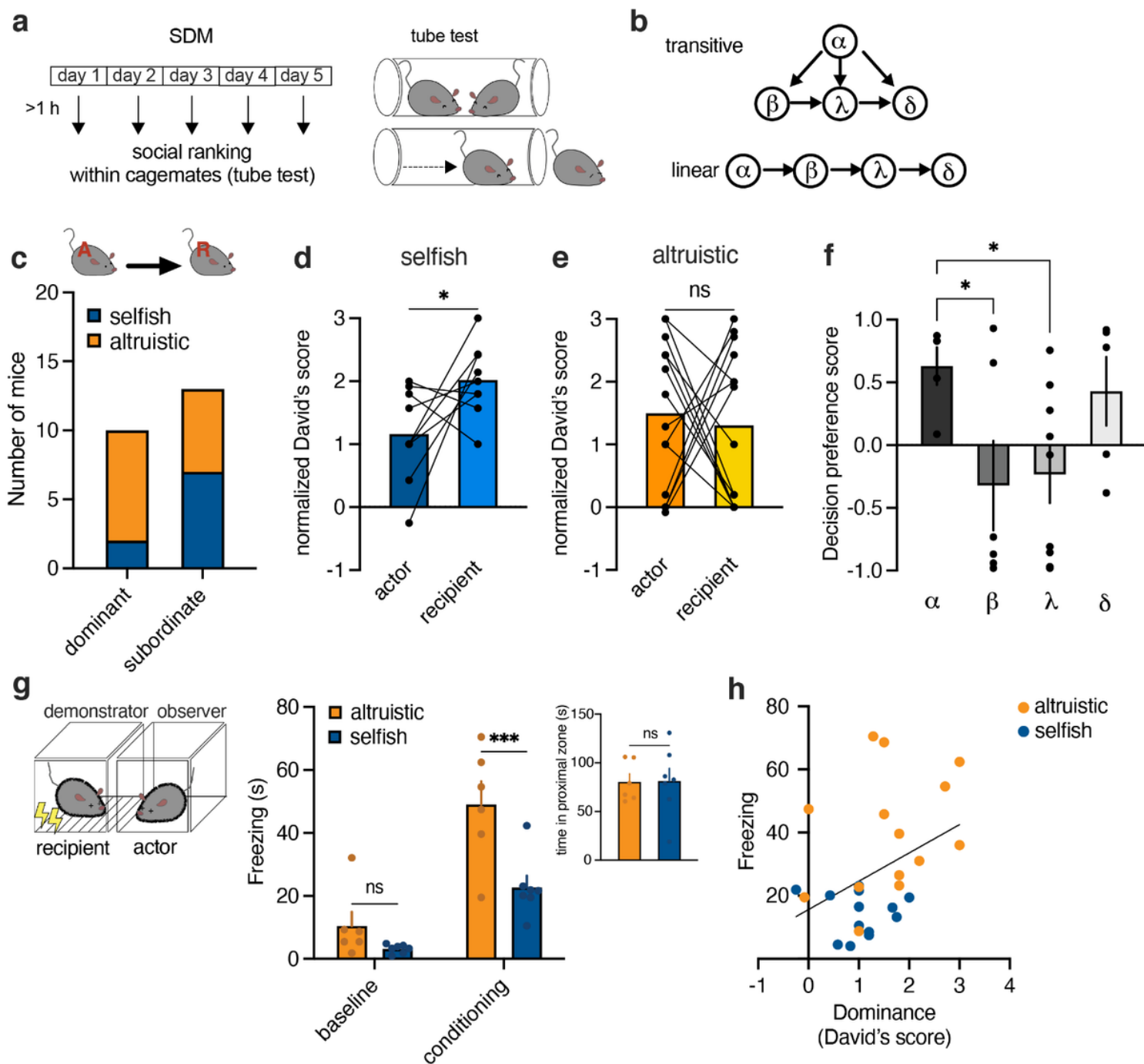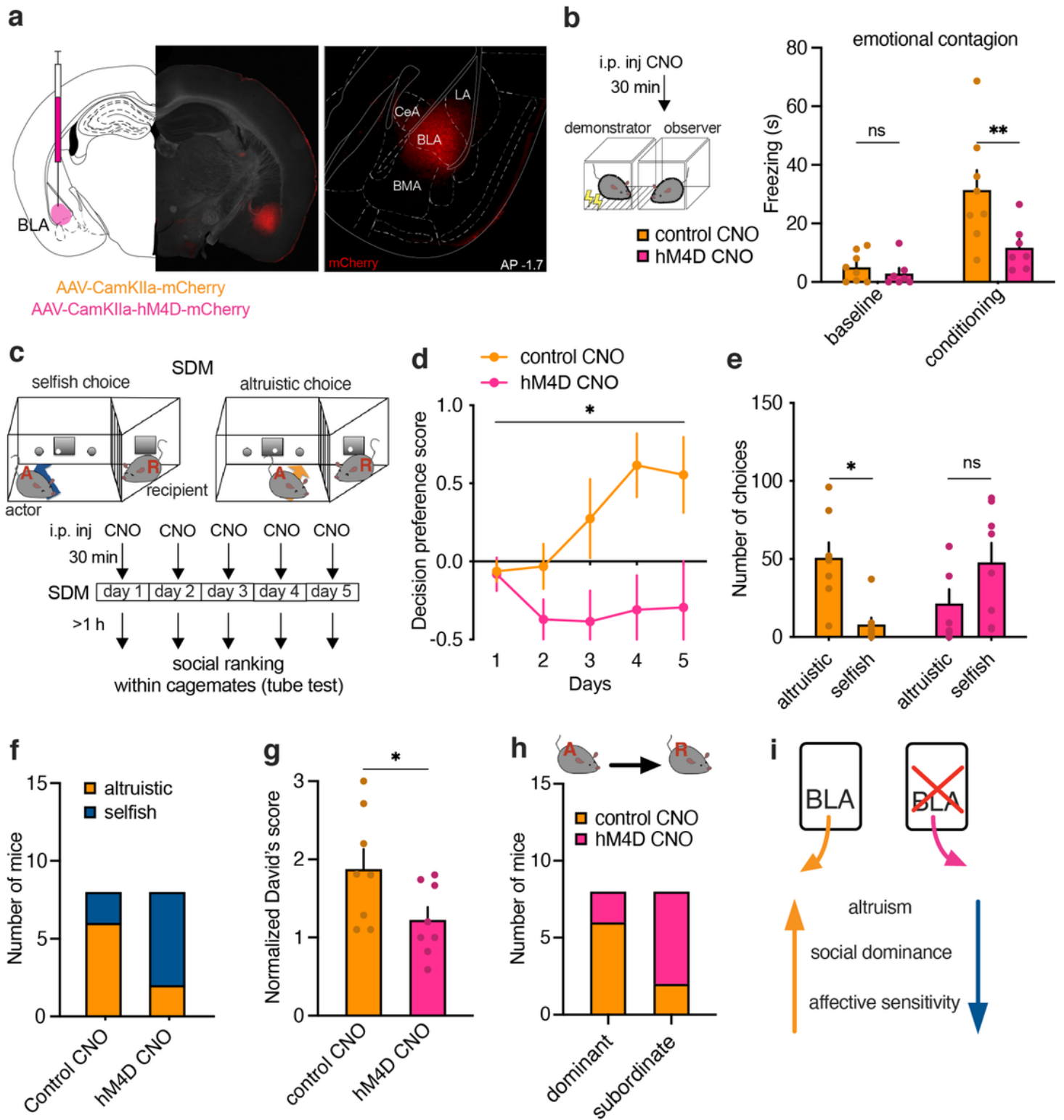
Figure 4

Social dominance hierarchy modulates preference for altruistic choices. a, After SDM daily session mice were tested on the tube test (at least 1h after SDM), to measure the hierarchical relationship of animals within the same cage. Actor and recipient mice were tested pairwise and using a round robin design. b, in all tested cages (n=8) the relation between animals was transitive and linear. c, number of dominant or subordinate actor mice compared to their recipient conspecific (n=23; Fisher's exact test p=0.11). (d-e), social dominance (normalized David's Score) quantified based on the number and directionality of interactions in the tube test in actor and recipient mice grouped by selfish (d, two-tailed paired t-test: t=2.72, d.f.=16, p=0.0151) and altruistic (e, t=0.43, d.f.=26, p=0.6698) actors and respective recipient

conspecific. f, decision preference score in mice grouped by social rank (one-way ANOVA, $F_{(3, 21)}=2.94$, $p=0.0567$; two-tailed paired t-test, alpha vs. beta: $t=2.28$, d.f.=9, $p=0.0478$; alpha vs. gamma: $t=2.64$, d.f.=12, $p=0.0215$). g, Left, Schematic representation of the observational fear learning. Right, freezing behavior in actor mice, grouped by altruistic or selfish preference during baseline and conditioning phases of the test (two-way RM ANOVA, phase (baseline, conditioning) x group (altruistic, selfish), $F_{(1, 11)}=6.23$, $p=0.0293$) and total time spent in the zone in the proximity of the divider between the actor and recipient compartment during the conditioning (two-tailed paired t-test: $t=0.04$, d.f.=11, $p=0.9632$). h, social dominance (David's Score) predicts affective sensitivity (freezing behavior during emotional contagion) (linear regression n=27, $y=8.971x+15.61$, $F_{(1, 25)}=4.47$, $p=0.0446$). $*p<0.05$, $***p<0.001$. n.s. not significant. Values are expressed as mean ± s.e.m.

Figure 5

BLA neuronal silencing modulates preference for altruistic choices BLA through dominance hierarchy. a, male mice were bilaterally injected in the BLA with AAV-CamKIIa-mCherry (control CNO, orange) or AAV-CamKIIa-hM4D-mCherry (hM4D CNO, fuchsia). Representative image of a coronal section of BLA. b, observers mice received intraperitoneal (i.p.) injection of CNO (3 mg/kg) and after 30 minutes were tested with their respective demonstrators on the observational fear learning paradigm. Freezing behavior

displayed by actor mice (control CNO and hM4D CNO) during baseline and conditioning phases of the test (two-way RM ANOVA, phase (baseline, conditioning) x group (altruistic, selfish), $F_{(1, 13)}=4.37$, $p=0.05$). c, thirty minutes before daily SDM session with familiar recipients, control (control CNO) and BLA hM4D-injected (hM4D CNO) mice received i.p. injection of CNO. d, decision preference score in the five days of SDM in control CNO (n=8) and hM4D CNO (n=8) mice (two-way RM ANOVA, group (control CNO, hM4D CNO) x time (days 1-5), $F_{(4, 56)}=4.560$, $p=0.0029$; one sample t-test to chance (0.0), control CNO: t=2.859, df=39, p=0.0068; hM4D CNO: t=3.306, df=39, p=0.0020). e, number of choices (nose poke responses) associated with altruistic and selfish choices in control CNO and hM4D CNO mice (two-way RM ANOVA, group (control CNO, hM4D CNO) x response (altruistic, selfish), $F_{(1, 14)}=8.06$, $p=0.0131$). f, number of control CNO and hM4D CNO mice displaying preference for altruistic or selfish choices (n=16; Fisher's exact test p=0.0455). Mice were assigned to altruistic (orange) or selfish (blue) analyzing decision preference scores using one sample t-test to chance. g, At least 1 hour after SDM, all the mice were tested in the tube test for assessment of social ranking within the cages. social dominance (normalized David's Score) quantified based on the number and directionality of interactions in the tube test in actor mice grouped by control CNO and hM4D CNO (two-tailed paired t-test: t=2.15, d.f.=14, p=0.0493). h, number of dominant or subordinate actor mice compared to their recipient conspecific (n=16; Fisher's exact test p=0.13). i, schematic model of the involvement of the BLA in social decision making. *p<0.05, **p<0.01. n.s. not significant. Values are expressed as mean ± s.e.m.

Supplementary Files

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- SupplementaryInformation.docx