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Abstract

In mammals, positive tactile contact is recognized as an effective tool for triggering positive affective states. In this study, we investigated the benefits of providing automatic rotative brushes for group-housed horses. Our three aims were: i. to determine whether horses used automatic brushes and if so how they used them; ii. to investigate whether the presence of these brushes induced positive social behaviours (allogrooming), or aggressiveness, as can be observed when there is competition for a desired resource; iii to provide a preliminary explanation of the role of the positive facial expression displayed by some horses while being groomed.

Two brushes were installed in a large stable with free access to a paddock in which 40 horses were housed 7h30/day. For four days, video-cameras placed above the brushes continuously filmed the horses. First, analysis of the video footage demonstrated that brushes were used by almost 90% of the horses, mainly on areas that are not easily accessible to another horse during allogrooming, such as the head. Secondly, it revealed that among the horses that used the brush, 25.7% expressed positive social behaviour (allogrooming) at the same time, while none expressed aggressive behaviours. The brush thus seems to act as a catalyst for affiliative behaviours rather than competition for a desired resource. Thirdly, we observed their facial expression, especially a positive one, described when the horse are groomed (neck moderately raised, eyes open or half-closed, upper lip extended, ears turned backwards almost in line with the nose). More than half of the horses displayed this while using the brushes. Interestingly, when a horse expressed this face, it had a six times greater chance to subsequently start allogrooming with a conspecific. Several possible explanations are discussed, including a social function. Further research is needed to validate other criteria to help qualify whether or not this positive facial expression is a signal of intentional communication. From a practical point of view, the results show that the brushes provide both a direct benefit to the horses enabling them

to conduct self-grooming, and also in all likelihood, an indirect benefit by facilitating positive social interactions. This study encourages the installation of automatic brushes in places where horses are kept to improve their welfare and provide comfort.

Key Words: Equus caballus, positive affect, facial expression, emotional indicators, welfare, environmental enrichment

Highlights

- Automatic rotative brushes are used by almost 90% of horses observed
- They trigger positive behaviour (allogrooming) and no aggressiveness
- Allogrooming is more frequent if the horse previously displayed a positive facial expression
- This suggests a social function of this facial expression
- This study encourages the installation of automatic brushes to improve horse welfare

1. Introduction

In mammals, positive tactile contact is recognized as an effective tool for triggering positive affective states (Burgdorf and Panksepp, 2006). These positive effects of tactile contacts can be evaluated through changes in behaviour such as the expression of specific vocalizations, specific facial expressions, contact seeking, allogrooming (grooming between conspecifics) or a relaxed state in various species such as rodents (Burgdorf and Panksepp, 2006; Finlayson et al., 2016), sheep (Reefmann et al., 2009; Westerath et al., 2014; Tamioso et al., 2017), cattle (Schmied et al., 2008; Westerath et al., 2014), heifers (Bertenshaw and Rowlinson, 2008), dogs (Rehn et al., 2014) or horses (Feh and De Mazières, 1993; McBride et al., 2004; Mullard et al., 2017). Positive tactile contact also has physiological correlates in horses. It induces changes in parameters such as heart rate (Feh and De Mazières, 1993) or hormone levels, such as the basal oxytocin level (Lansade et al., 2018), for a review (VanDierendonck and Spruijt, 2012).

With the aim of improving welfare by promoting positive affective states, different ways of providing tactile contact have been studied in horses: allowing allogrooming with partners (Snorrason et al., 2003), a handler scratching them with their hands (Feh and De Mazières, 1993; Lansade et al., 2018), or by making different devices available, such as fixed brushes (Lansade et al., 2014). On cattle farms, automatically rotating brushes are also often provided and animals use them extensively (DeVries et al., 2007; Moncada et al., 2020) with positive results reported, such as a reduction in time spent inactive (Velasquez-Munoz et al., 2019) or an increase in self-grooming (Horvath and Miller-Cushon, 2019). To date, the influence of an automatic brush on the behaviour of group-housed horses has not been studied. Thus, the present study on horses aimed to investigate this.

Moreover, we recently showed in horses that positive tactile contacts trigger a positive affective state associated with a specific facial expression (called “positive facial expression” hereafter).

Indeed, when scratched by humans, horses express a facial expressions with the following characteristics: neck moderately raised, eyes opened or half closed, upper lip extended and either immobile or twitching and ears turned backwards almost in line with the nose (Lansade et al., 2018; Trösch et al., 2020). Facial expressions may have a double function: expressing passive emotion and/or an intention to engage in specific activities with the receiver (Waller et al., 2017; Camerlink et al., 2018). The communication of intention is thought to regulate social interactions within groups, such as lipsmacks in chimpanzees that enable allogrooming to be coordinated and prolonged, and facilitate social cooperation. According to Townsend et al. (2017), to determine whether facial expressions are intentional at least two criteria should be used to evaluate them: the signal is only produced in the presence of the recipient (audience effect) and it is contingent on the behaviour of the recipient, which means that the behaviour follows the facial expression and -de facto- ends it (audience behaviour). The intentional use of facial expressions in social communication remains highly debated (Waller et al., 2017) and to date has not been explored in horses.

This study had three aims. The first was to determine whether or not horses used automatic brushes, on what parts of their body, at what frequency, and how they triggered them. The second aim was to investigate whether the presence of these brushes induced positive social behaviours assessed by the expression of allogrooming behavior, or instead negative behaviours such as threats and aggressiveness, as can be observed when there is competition for a desired resource. The third aim was to provide a preliminary description of the role of facial expressions in horses, by determining whether the positive facial expression described when horses are groomed by humans was also expressed when they used the brush, and whether it played a social role or not. To this end, we investigated whether there was an audience effect when the positive face was expressed (i.e. if a conspecific was nearby when the signal was produced) and audience behaviour in response (with a contingent signal to the recipient's behaviour). We

97 expected that the positive facial expression would be expressed while brushing but
98 preferentially when another horse was nearby and that this facial expression would promote
99 allogrooming.

100

101 ## 2. Material and Method

102

103 ### 2.1. Welfare statement

104 This study was non-invasive: we filmed the animals in their normal living condition without
105 particular intervention. No permission from the Animal Ethics Committee was needed.

106

107 ### 2.2. Animals, housing and characteristics of the brushes

108 The study was conducted on 40 female Welsh ponies, with an average age of 7.56±0.42
109 (mean±sem) years old and height at withers of 1m to 1.20m, reared at the Animal Physiology
110 Experimental Unit PAO, INRAE (DOI: 10.15454/1.5573896321728955E12). The herd was
111 established four weeks before the beginning of the observations. From then on, they lived in a
112 group at pasture and every day from 08:00 to 15:30 they were brought into a large straw-covered
113 stabling area (11x35m) with free access to an adjoining paddock (10x35m). Hay and water were
114 available ad libitum in the stabling area.
115 Two brushes were present in the middle of the stabling area 10m apart. The brushes were those
116 usually used for goats and which start rotating when touched by an animal (Mini rotating brush
117 for goats and calves, MSB DeLaval). Each brush was attached to a mobile axis, enabling it to
118 move from a vertical to a horizontal position and thus reach different parts of the pony's body
119 (Fig.1). The top of the brush was at a height of 120cm, corresponding approximately to the

120 pony's' withers. The dimensions of each brush were 72x30 cm and the rotation speed was 25.5
121 rotations/min.
122
123 **2.3. Behavioural observations**
124 After 4 weeks of familiarization to the presence of the brushes in the stabling area, a camera
125 was fixed above each brush to film the horses using it. Four periods from 08:00 to 15:30 were
126 recorded, two in week 5 (14, 15 May) and two in week 6 (22, 23 May), for a total of 30 hours
127 of observation. The film footage was subsequently viewed by an experienced observer (JL) to
128 record the following behaviours which were divided into three categories according to the
129 different aims.
130 To characterize the use of the automatic brushes (aim 1), we recorded: the number of sequences
131 when a horse activated the brush, and identified the animal; the time the brush was used for
132 each sequence (i.e. the time the brush was in contact with the horse); the part of the body that
133 activated the brush (head, forehand or hindquarters, Fig.3) or if it was already activated when
134 the horse arrived. The percentage of time spent brushing each of these areas (time on one area
135 of the body/ total time of use*100) was also calculated.
136 Then, we determined what type of behaviour was performed during the brushing (aim 2). For
137 each sequence, we recorded whether at the same time as the horse used the brush it
138 demonstrated an affiliative behaviour such as allogrooming (*the horse is top to tail with a*
139 *conspecific and both mutually nibbling different parts of each other's bodies*) or an aggressive
140 behaviour such as threatening to bite and biting *(ears are backward, neck and head are*
141 *stretched toward the opponent and accompanied by a quick movement toward it, without the*
142 *teeth touching the opponent - biting is similar, but the teeth touch the opponent)*, threatening to
143 kick and kicking *(the ears are backward, the croup is toward the opponent and the horse raises*
144 *one or both hind limbs or kicks out the hind limbs - kicking is similar, but the limbs touch the*

145 *opponent)*, chasing or attacking. For each sequence, this was recorded as a binary response:
146 Yes/No.

147 For each sequence, we also evaluated the facial expressions of the horses while they used the
148 brush (aim 3): a positive facial expression (first described during grooming by a person, see
149 Lansade et al., 2018), a negative facial expression, or neither of them (see Fig.2 for a description
150 of each expression). The negative facial expression differs from the threatening behaviors as
151 the former is not explicitly directed toward a conspecific (the head and neck are not stretched
152 toward a specific opponent and it is not accompanied by a quick movement toward it; the horse
153 does not raise or kick out its hind limbs). When the four zones used to describe the facial
154 expressions were not entirely visible (neck, ears, eyes and lips) the sequence was excluded from
155 the analyses. Only one of these facial expressions was ever observed in any one sequence of
156 brush use, never both of them. Finally we noted whether the positive facial expressions were
157 expressed when there was another horse within 2m (Yes/No) and whether it was followed by
158 allogrooming that *de facto* ended the facial expression (Yes/No). Initially, we had planned to
159 do this kind of analysis for the negative facial expression to determine whether it was also
160 followed by the aggressive behaviours, but since aggressive behaviours were not expressed,
161 this was not possible.

162

163 **2.4. Statistical Analysis**

164 Two-proportion z tests using the Monte Carlo Method (5000 simulations, Fleiss et al., 2003;
165 Vose, 2008) were conducted to compare the proportion of affiliative behaviours vs aggressive
166 behaviours; to compare the proportion of positive vs negative facial expressions; and to test
167 whether positive facial expressions were associated with affiliative behaviours (allogrooming).

For the latter, we compared the proportion of allogrooming after a positive facial expression vs that without a positive facial expression.

We observed 139 sequences during which a horse used the brush. Of the 139 times the brushes were used, it was possible to determine 132 times when allogrooming took place or not, 111 times when there was a positive facial expression or not and 131 times when there was a negative facial expression or not. These numbers differed since it was not always possible to validate all the criteria presented in Fig. 2. For instance, sometimes it was not possible to see all the criteria required to validate the presence of a positive facial expression, because for example the lips were not visible on the video, and therefore their position was unclear. In that case, a positive facial expression could not be validated and we considered the data as missing. The two variables 'allogrooming' and 'positive facial expression' could be matched for 105 sessions of brush use. The statistical analyses presented in the results section were conducted on these respective numbers of sequences.

3. Results

3.1. Descriptive analysis of the use of the automatic brushes

Over the four 08:00 to 15:30 periods, 35 of the 40 horses used a brush (i.e. 87.5%) and the brushes were activated 139 times. The mean $\pm$ sem for each horse was 3.97 ± 2.93 times, with a minimum of once and a maximum of 11 times per horse. The mean $\pm$ sem duration of use was 55.28 ± 4.125 s. Of the 139 times the brush was activated, the majority were triggered by the horse's head, followed by the forehand and then the hindquarters. In a quarter of the cases the brush was already activated when the horse arrived (Fig.3a). The horses mainly scratched their heads, followed by their hindquarters and then their forehand (Fig.3b).

3.2. Aggressive and affiliative behaviours resulting from the use of the brushes

There were no cases of aggressive behaviour when horses used the brush. By contrast, affiliative behaviour (allogrooming) was expressed in 7.57% of the sequences, by nine different horses - one horse allogroomed twice (comparison between the proportion of aggressive behaviour and affiliative behaviour: 0/139 vs 10/132 sequences, two-proportion z test using the Monte Carlo Method, diff=0.076, p= 0.001).

3.3. Facial expressions observed while the horse used the brush and the social context in which they appeared

There were only 3.82% of sequences with a negative facial expression (5/131 sequences, from 5 different horses), but 39.94% with a positive facial expression (41/111 sequences, from 22 different horses, two-proportion z test using the Monte Carlo Method, diff=0.33, p<0.0001, Fig.4).

Regarding the 'audience effect' hypothesis, there was always at least one other horse present less than 2 m from the horse being brushed with the exception of one case (and in that case, the horse did not produce a specific facial expression), thus it was not possible to test this hypothesis.

Regarding the 'audience behaviour' hypothesis, among the 105 sequences for which the variables "allogrooming" and "positive facial expression" matched, horses displayed a facial expression in 37 sequences and did not in 68 sequences. Interestingly, when the horse expressed a positive facial expression during a sequence there was 18.91% chance that an allogrooming event with another horse would take place immediately after it (7/37 sequences, for 7 different horses). This was 6.4 times less if the horse had not previously made a positive facial expression

(2.94%: 2/68 sequences, for 2 different horses). These proportions are significantly different (two-proportion z test using the Monte Carlo Method, diff=-0.16, p=0.008, Fig. 5). The 9 (7+2) sequences of allogrooming involved nine different horses being brushed. We can also notice that allogrooming was never observed after a horse had expressed a negative facial expression (n=5).

4. Discussion

This study first demonstrates that automatic rotating brushes were used by a large proportion of horses living in a group. It also revealed that when horses used these brushes more positive social behaviours (allogrooming) than aggressive behaviours were expressed. Finally, if a horse expressed a positive facial expression during a sequence, it had a six times greater chance to subsequently start allogrooming with a conspecific than if it did not.

4.1. Description of brush use

Like cows (e.g. DeVries et al., 2007; Horvath and Miller-Cushon, 2019; Velasquez-Munoz et al., 2019; Moncada et al., 2020), horses spontaneously use automatic brushes, as 87% of the horses in this study used them during the four 7h30 observation periods. The duration of each use was generally short being less than a minute, but this corresponds to the durations described in other species, for instance, bout duration in calves was 18 s (Zobel et al., 2017). The number of visits to the brushes (4 visits per animal over 30h, i.e. 3.2 visits / 24h) is also on the same scale as that observed in adult cows : 4.5 visits / 24h in Mandel et al. (2013) or 7.7 visits / 24h in DeVries et al. (2007). This figure is also similar to that described in Islandic horses for allogrooming sequences (Snorrason et al., 2003).

240 Regarding the areas brushed the most, we might have expected to find the base of the neck or
241 the withers, which according to a study on Camargue horses corresponds to the area of the body
242 that horses mutually scratch during allogrooming (Feh and De Mazières, 1993). However, this
243 was not the case in our study, as the horses spent more time scratching their head and
244 hindquarters than the base of the neck which was nevertheless an area easily accessible to the
245 automatic brush. In particular, the head is not an area that horses mutually groom probably
246 because it is not so easy for them, but it was the area brushed the most by the automatic brush.
247 The brushes thus appear to supplement the allogrooming horses carry out. That could be
248 particularly true in spring and summer, when potential itching due to flies might explain why
249 horses use the brushes more on areas not typically associated with allogrooming (the present
250 observations were made in May). The time spent using the brush could also reflect a tendency
251 to play with them, although this hypothesis cannot be verified.

252 To activate the brush, horses mainly used their head and their forehand. However, it should be
253 noted that in a quarter of the cases they used brushes that had already been activated by another
254 horse. It is possible that this was based on a process of social facilitation ("when the behaviour
255 of a conspecific changes the motivation of the observer, resulting in the tendency of individual
256 animals to do what other individuals are doing") or local enhancement ("when the behaviour of
257 the demonstrator results in an increase in the salience of a particular stimulus, and the observer's
258 motivation to investigate that stimulus may be increased"),processes previously described in
259 the horse (Rørvang et al., 2018). The horses could thus have used the brush because a
260 conspecific had just previously used it. This raises the question of the role these brushes could
261 play in social cohesion.

262

263 **4.2. Induction of affiliative behaviours**

The most striking result of this study is that the brushes induced more affiliative than aggressive behaviours. This is noteworthy as we could have expected the contrary regarding a limited resource with only two brushes for 40 horses. Indeed, there could have been a form of competition to access this resource, with aggressive behaviours of a threatening or biting type, as can be observed regarding a vital and rare source, such as at a watering place (Rutberg and Greenberg, 1990). No such behaviour was observed. On the contrary, we were surprised to note that while horses were being brushed by the automatic brush they simultaneously started allogrooming. The brush thus seems to act as a catalyst for affiliative behaviour. To confirm this, in a future study it would be interesting to count the total number of these affiliative behaviours in the group of horses that can access the brushes compared to groups of horses that cannot. In any case, our finding suggests that the presence of brushes directly improved horse welfare by providing them with comfort, as evidenced by the presence of positive facial expression in almost half of the brushing sequences, but also indirectly, by fostering positive social relations between the horses, which is essential to maintain social cohesion (review: VanDierendonck and Spruijt, 2012).

4.3. Social context of expressing the positive facial expression

Finally, this study provides initial leads into understanding the social role of horses facial expressions, and in particular the positive facial expression observed when horses are groomed (Lansade et al., 2018). First, we aimed to investigate whether there was an audience effect (if a conspecific was nearby when the facial expression was produced). However, this effect could not be tested because in all sequences except one, there was a horse less than 2m from the brushed horse. Second, our aim was to investigate whether there was audience behaviour in response. Results show that if a horse expressed this face during the brushing sequence (half of the horses were observed doing this), it had a six times greater chance to subsequently start

allogrooming with a conspecific than if it had not expressed that face. There are at least three possible explanations of this result.

The first corresponds to the audience behaviour hypothesis: the positive facial expression could promote allogrooming and could be used intentionally to communicate. However, to confirm this explanation further studies are necessary to identify supplementary criteria. Among the criteria put forward by Townsend et al. (2017), there is the "Manipulation of the attentional state of the recipient". To investigate this, we could determine whether "attention-getting behaviours were directed towards a recipient" before the facial expression was expressed (Liebal et al., 2004). We could also check whether there was an exchange of gazes between the signaler and the recipient, given that in horses, gaze alternations to manipulate the attention of the recipient have already been described in another context (Malavasi and Huber, 2016). In our study, there were too many horses in front of the brushed horse to be able to distinguish clearly this type of behaviour. Moreover, as it is known that horses preferentially display allogrooming with certain privileged partners (review: VanDierendonck and Spruijt, 2012), it could also be interesting to determine the composition of the audience, and investigate whether this potential signal was only produced in the presence of certain recipients.

A second explanation is related to a phenomenon of emotional contagion. In a previous experiment, we showed that horses tended to engage in allogrooming toward an experimenter simply by watching video footage of another horse expressing this positive facial expression while it was scratched by a handler (Trösch et al., 2020). This behaviour was interpreted as a phenomenon of positive emotional contagion. To discriminate between intentional communication and emotional contagion, we could try to determine whether the positive facial expression of horses using the brush specifically promotes allogrooming between these particular horses and privileged partners, or also allogrooming between two third-party horses (who are not using the brushes).

Finally, a third explanation is that the animals who displayed the positive facial expression could be those which appreciated being scratched by the brush the most, and which were the most motivated to be groomed whether with a brush or by a conspecific. In this case they could also be the most motivated to subsequently start allogrooming with another horse.

In any case, our result shows that the brush could act as a catalyst for positive emotions and affiliation behaviours within the group.

5. Conclusions

This study shows that 87.5% of the horses observed used the brushes. The automatic brushes provide both a direct benefit to the horses enabling them to conduct self-grooming, and also in all likelihood, an indirect benefit by facilitating positive social interactions (allogrooming). They do not trigger aggressiveness, as can be sometimes observed when there is competition for a desired resource. The brush thus would appear to act as a catalyst for affiliative behaviours. These results promote the installation of automatic brushes in places where horses are kept to improve their welfare and provide comfort. Our results also show that more than half of the horse expressed a positive facial expression while using the brush. When they displayed this facial expression, they had a six times greater chance to subsequently start allogrooming with a conspecific than if they did not. For a clearer interpretation of this result further research is needed to validate other criteria to qualify whether or not this positive face is a signal of intentional communication.

References

Bertenshaw, C.E., Rowlinson, P., 2008. Exploring heifers' perception of 'positive' treatment through their motivation to pursue a retreated human. Anim. Welfare 17, 313-319.

Burgdorf, J., Panksepp, J., 2006. The neurobiology of positive emotions. Neurosci. Biobehav. Rev. 30, 173-187. doi:http://dx.doi.org/10.1016/j.neubiorev.2005.06.001
Camerlink, I., Coulange, E., Farish, M., Baxter, E.M., Turner, S.P., 2018. Facial expression as a potential measure of both intent and emotion. Sci Rep 8, 17602. doi:10.1038/s41598-018-35905-3
DeVries, T.J., Vankova, M., Veira, D.M., von Keyserlingk, M.A.G., 2007. Short communication: Usage of mechanical brushes by lactating dairy cows. J. Dairy Sci. 90, 2241-2245. doi:10.3168/jds.2006-648
Feh, C., De Mazières, J., 1993. Grooming at a preferred site reduces heart rate in horses. Anim. Behav. 46, 1191-1194.
Finlayson, K., Lampe, J.F., Hintze, S., Wurbel, H., Melotti, L., 2016. Facial Indicators of Positive Emotions in Rats. PLoS ONE 11. doi:10.1371/journal.pone.0166446
Fleiss, J., L. , Bruce, L., Myunghee, C.P., 2003. Statistical Methods for Rates and Proportions. Third Edition ed. John Wiley & Sons. doi:10.1002/0471445428
Horvath, K.C., Miller-Cushon, E.K., 2019. Characterizing grooming behavior patterns and the influence of brush access on the behavior of group-housed dairy calves. J. Dairy Sci. 102, 3421-3430. doi:10.3168/jds.2018-15460
Lansade, L., Nowak, R., Lainé, A.-L., Leterrier, C., Bonneau, C., Parias, C., Bertin, A., 2018. Facial expression and oxytocin as possible markers of positive emotions in horses. Sci Rep 8, 14680. doi:10.1038/s41598-018-32993-z
Lansade, L., Valenchon, M., Foury, A., Neveux, C., Cole, S.W., Laye, S., Cardinaud, B., Levy, F., Moisan, M.-P., 2014. Behavioral and Transcriptomic Fingerprints of an Enriched Environment in Horses (Equus caballus). PLoS ONE 9. doi:10.1371/journal.pone.0114384
Liebal, K., Call, J., Tomasello, M., 2004. Use of gesture sequences in chimpanzees. Am. J. Primatol. 64, 377-396. doi:10.1002/ajp.20087
Malavasi, R., Huber, L., 2016. Evidence of heterospecific referential communication from domestic horses (Equus caballus) to humans. Anim Cogn 19, 899-909. doi:10.1007/s10071-016-0987-0
Mandel, R., Whay, H.R., Nicol, C.J., Klement, E., 2013. The effect of food location, heat load, and intrusive medical procedures on brushing activity in dairy cows. J. Dairy Sci. 96, 6506-6513. doi:https://doi.org/10.3168/jds.2013-6941
McBride, S.D., Hemmings, A., Robinson, K., 2004. A preliminary study on the effect of massage to reduce stress in the horse. J Equine Vet Sci 24, 76-81. doi:10.1016/j.jevs.2004.01.005
Moncada, A.C., Neave, H.W., von Keyserlingk, M.A.G., Weary, D.M., 2020. Use of a mechanical brush by dairy cows with chorioptic mange. Appl. Anim. Behav. Sci. 223, 104925. doi:https://doi.org/10.1016/j.applanim.2019.104925
Mullard, J., Berger, J.M., Ellis, A.D., Dyson, S., 2017. Development of an ethogram to describe facial expressions in ridden horses (FEReq). Journal of Veterinary Behavior-Clinical Applications and Research 18, 7-12. doi:10.1016/j.jveb.2016.11.005
Reefmann, N., Wechsler, B., Gygax, L., 2009. Behavioural and physiological assessment of positive and negative emotion in sheep. Anim. Behav. 78, 651-659. doi:http://dx.doi.org/10.1016/j.anbehav.2009.06.015
Rehn, T., Handlin, L., Uvnas-Moberg, K., Keeling, L.J., 2014. Dogs' endocrine and behavioural responses at reunion are affected by how the human initiates contact. Physiol. Behav. 124, 45-53. doi:10.1016/j.physbeh.2013.10.009
Rørvang, M.V., Christensen, J.W., Ladewig, J., McLean, A., 2018. Social Learning in Horses—Fact or Fiction? Frontiers in Veterinary Science 5. doi:10.3389/fvets.2018.00212
Rutberg, A.T., Greenberg, S.A., 1990. Dominance, aggression frequencies and modes of aggressive competition in feral pony mares. Anim. Behav. 40, 322-331. doi:https://doi.org/10.1016/S0003-3472(05)80927-3
Schmied, C., Waiblinger, S., Scharl, T., Leisch, F., Boivin, X., 2008. Stroking of different body regions by a human: Effects on behaviour and heart rate of dairy cows. Appl. Anim. Behav. Sci. 109, 25-38. doi:10.1016/j.applanim.2007.01.013

389 Snorrason, S., Sigurjónsdóttir, H., Thórhallsdóttir, A., van Dierendonck, M., 2003. Social relationships
390 in a group of horses without a mature stallion. Behaviour 140, 783-804.
391 doi:https://doi.org/10.1163/156853903322370670
392 Tamioso, P.R., Rucinque, D.S., Taconeli, C.A., da Silva, G.P., Molento, C.F.M., 2017. Behavior and body
393 surface temperature as welfare indicators in selected sheep regularly brushed by a familiar observer.
394 J Vet Beh 19, 27-34. doi:https://doi.org/10.1016/j.jveb.2017.01.004
395 Townsend, S.W., Koski, S.E., Byrne, R.W., Slocombe, K.E., Bickel, B., Boeckle, M., Braga Goncalves, I.,
396 Burkart, J.M., Flower, T., Gaunet, F., Glock, H.J., Gruber, T., Jansen, D.A.W.A.M., Liebal, K., Linke, A.,
397 Miklósi, Á., Moore, R., van Schaik, C.P., Stoll, S., Vail, A., Waller, B.M., Wild, M., Zuberbühler, K.,
398 Manser, M.B., 2017. Exorcising Grice's ghost: an empirical approach to studying intentional
399 communication in animals. Biological Reviews 92, 1427-1433. doi:https://doi.org/10.1111/brv.12289
400 Trösch, M., Pellon, S., Cuzol, F., Parias, C., Nowak, R., Calandreau, L., Lansade, L., 2020. Horses feel
401 emotions when they watch positive and negative horse–human interactions in a video and transpose
402 what they saw to real life. Anim Cogn. doi:10.1007/s10071-020-01369-0
403 VanDierendonck, M.C., Spruijt, B.M., 2012. Coping in groups of domestic horses – Review from a
404 social and neurobiological perspective. Appl. Anim. Behav. Sci. 138, 194-202.
405 doi:https://doi.org/10.1016/j.applanim.2012.02.007
406 Velasquez-Munoz, A., Manriquez, D., Paudyal, S., Solano, G., Han, H., Callan, R., Velez, J., Pinedo, P.,
407 2019. Effect of a mechanical grooming brush on the behavior and health of recently weaned heifer
408 calves. BMC Vet. Res. 15. doi:10.1186/s12917-019-2033-3
409 Vose, D., 2008. Risk Analysis - A Quantitative Guide. Third Edition ed. John Wiley & Sons, New York.
410 Waller, B.M., Whitehouse, J., Micheletta, J., 2017. Rethinking primate facial expression: A predictive
411 framework. Neurosci. Biobehav. Rev. 82, 13-21. doi:https://doi.org/10.1016/j.neubiorev.2016.09.005
412 Westerath, H.S., Gygax, L., Hillmann, E., 2014. Are special feed and being brushed judged as positive
413 by calves? Appl. Anim. Behav. Sci. 156, 12-21. doi:http://dx.doi.org/10.1016/j.applanim.2014.04.003
414 Zobel, G., Neave, H.W., Henderson, H.V., Webster, J., 2017. Calves Use an Automated Brush and a
415 Hanging Rope When Pair-Housed. Animals (Basel) 7, 84. doi:10.3390/ani7110084

Declaration of Competing Interest

The authors report no declarations of interest.

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Figures captions

Figure 1: Automatic brush being used by a horse in the study

Figure 2. Behaviour and facial expressions recorded while the horse used the brush

Figure 3 a. Parts of the body used to activate the brush (as a percentage of the 139 activations) and **b**. percentage of time spent brushing each of these different parts.

Figure 4. Affiliative or aggressive behaviours and facial expressions observed while the horse used the brush**.**

***p<0.001

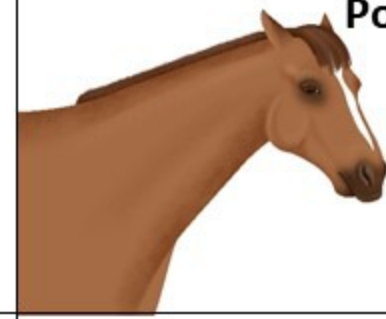
Data are expressed as a percentage of sequences during which the behaviour or the facial expression was observed

Figure 5. Percentage of chance that the horse using the brush simultaneously allogroomed another horse, when there was or not a prior positive facial expression

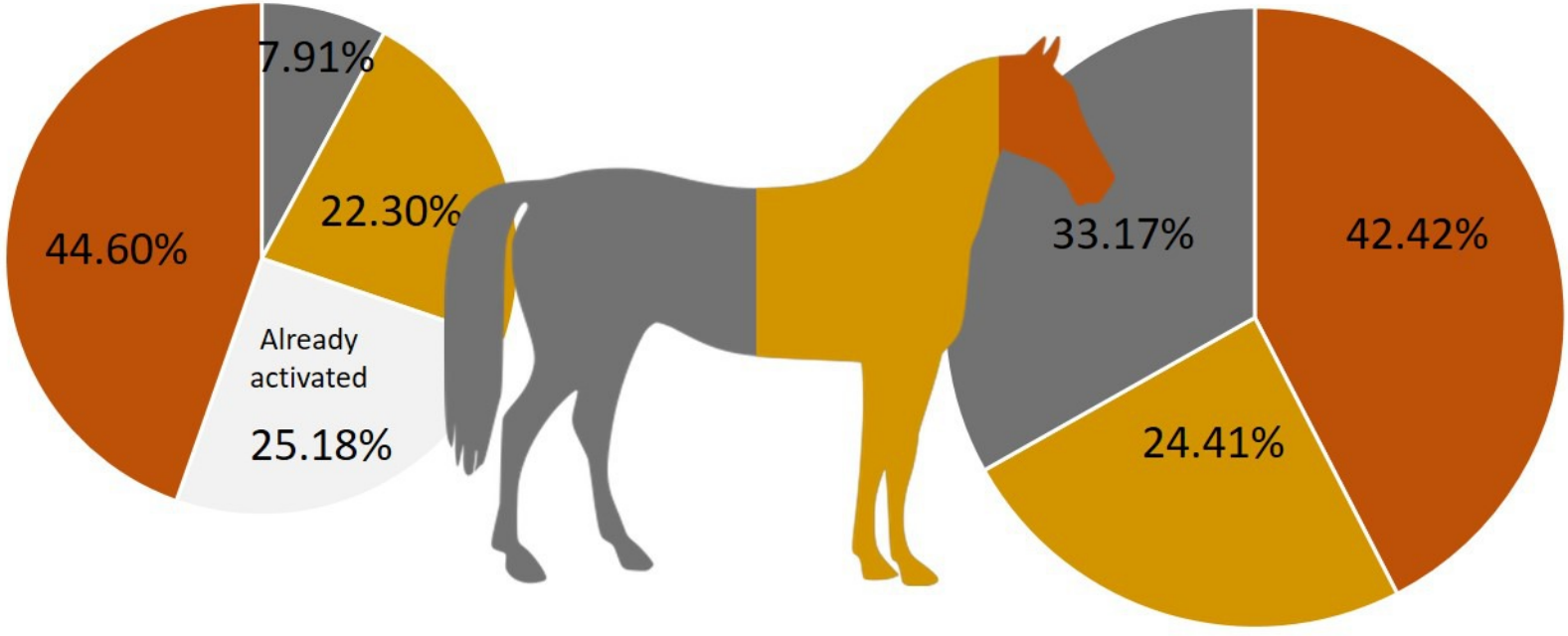
**p<0.01

 : presence or absence of a positive facial expression during the sequence

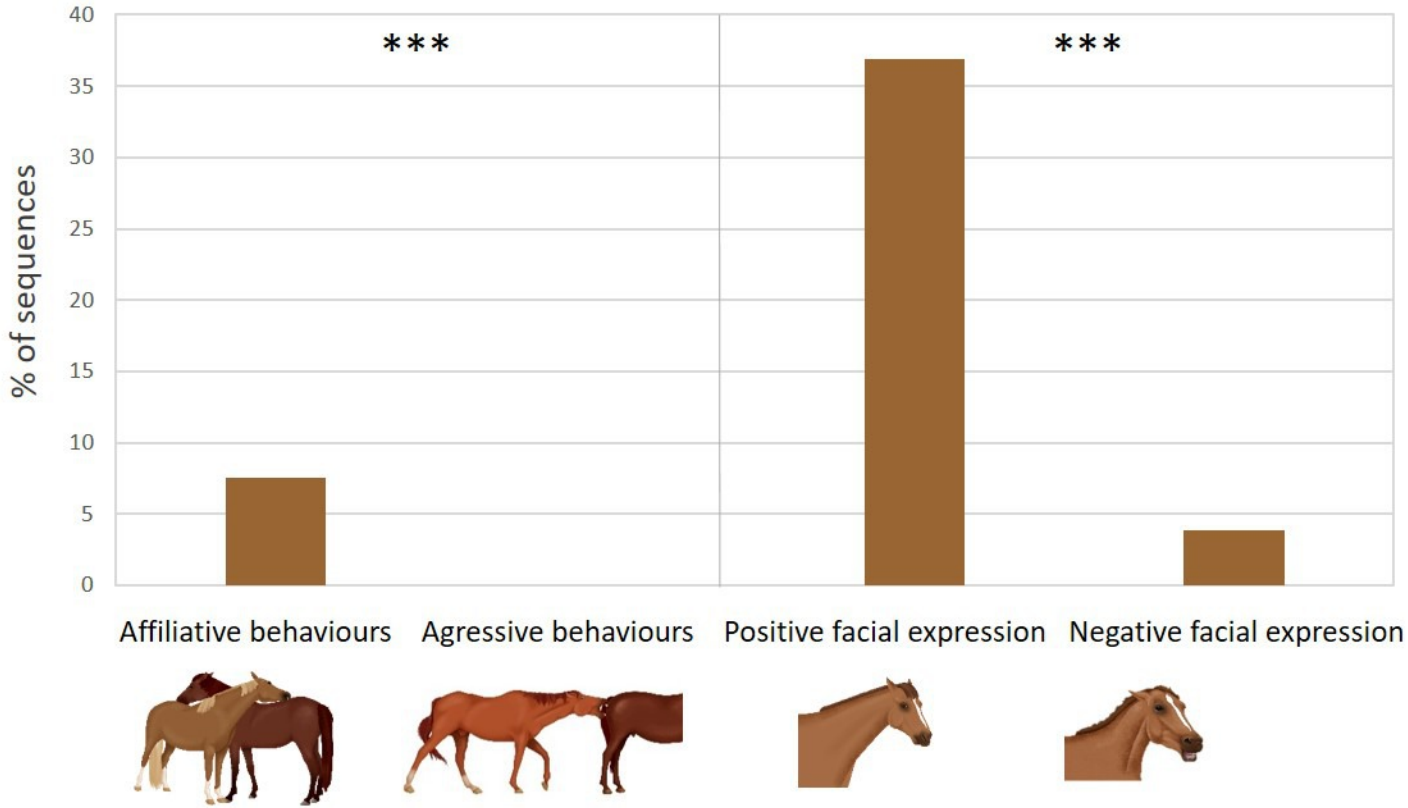
 : allogrooming during the sequence

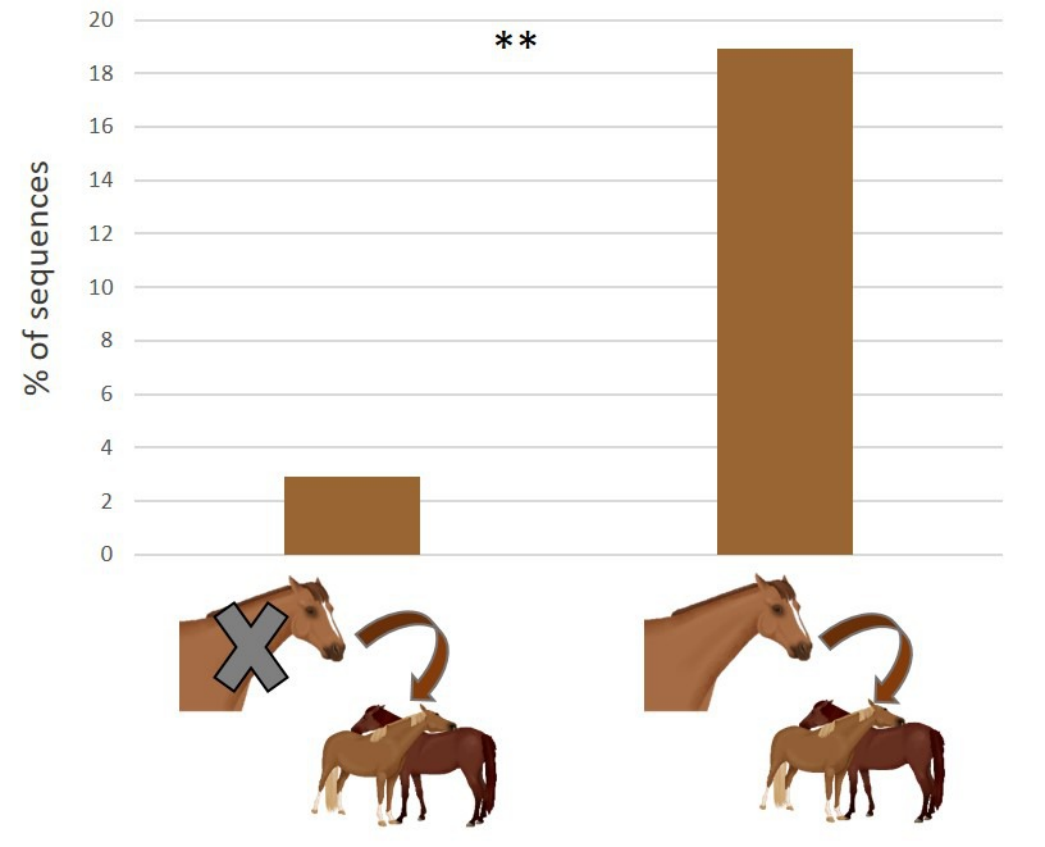
Behaviours	Facial expressions
Affiliative behaviour  *Allogrooming*	**Positive facial expression**  *Upper lip extended forward* *Relaxed nostrils (not dilated)* *Eyes opened to half closed* *Ears turned backward* *Neck moderately raised*
Agressive behaviour  *Threatening to bite* *Threatening to kick* *Biting, Kicking* *Chasing, Attacking*	**Negative facial expression**  *Angle head/neck opened* *Ears pinned back in the mane* *Eyes wide-open* *Optional: Mouth open/teeth visible*

a. Part of the body used to activate the brusch



b. Body part brushed


40
35
30
25
20
15
10
5
0
% of sequences
***
***
Affiliative behaviours
Agressive behaviours
Positive facial expression
Negative facial expression


20
18
16
14
12
10
8
6
4
2
0
% of sequences
**