

1       **A    NOTE    ON    REACTION    TO    NOVEL**  
2       **STIMULUS        AND        RESTRAINT        BY**  
3       **THERAPEUTIC RIDING HORSES**

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1 **Abstract**

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3 Little research has been done to measure reactivity objectively in therapeutic riding horses  
4 (TRH). As individual reactivity and chronic stress could be assessed by exposing animals to  
5 acute, novel stressors, the authors of this work aimed at comparing reactions of TRHs and  
6 jumping horses (JH) to two challenges. Four TRHs and four JHs were exposed to a restraint  
7 covering their head with a hood for 1 hour and to a startling stimulus (a 40 cm long, red and  
8 white synthetic holiday garland shaken with a rustling noise inside the box). Heart rate (HR)  
9 and heart rate variability (HRV) were recorded continuously and telemetrically, the reaction  
10 was video-recorded and analysed with a software for behavioural analysis. Blood samples  
11 were collected before and after each challenge to determine lymphocyte proliferation and  
12 other biochemical parameters. Horses spent most of the time immobile during the challenges  
13 ( $p < 0.05$ ). TRHs had a significantly higher average basal HR than JH ( $p < 0.05$ ), probably due  
14 to their better condition. HR varied among different behaviours during the restraint ( $p < 0.05$ ):  
15 the average HR during "pawing" was higher than during other behaviours ( $p < 0.005$ ). A  
16 significant decrease in the proliferation of lymphocytes in samples taken after the removal of  
17 the hood ( $p < 0.05$ ) was found, while the other stress related parameters didn't vary  
18 significantly after the challenges. The authors conclude that TRHs did not react less than JHs  
19 to the new stimuli and this should be taken into consideration while planning their daily work  
20 and management.

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23 **Keywords:** horse, therapeutic riding, behavioural indicators, heart rate, lymphocyte  
24 proliferation.

1 **1. Introduction**

2

3 Therapeutic riding (TR) is an animal-assisted therapy supporting or reinforcing motor,  
4 bio-chemical and mental functions in disabled people by means of horse-riding (De Lubersac  
5 and Lallery, 1977). Physical and behavioural characteristics, health conditions and  
6 management patterns associated with the horses being used in such therapy are critical to the  
7 program's quality as well as its success. In terms of reactivity, a therapeutic riding horse  
8 (TRH) should not be excessively reactive. Nor should it be excessively reactive to new  
9 stimuli or prone to sudden movements (Engel, 1992). Any reactivity variation in horses to  
10 behavioural tests may indicate the existence of stressful situations (Visser, 2001). If  
11 neglected, they might turn out to be a potential cause of problems for the horse's well-being  
12 as well as for the patients safety. So far few studies have been published aiming at measuring  
13 TRHs' behaviour and reactivity objectively as well as assessing whether these features might  
14 be used as selection criteria (Anderson et al., 1999). Furthermore, few researches have been  
15 carried out in order to emphasise whether the work performed by TRHs might negatively  
16 impact their well-being and, as a consequence, therapy quality. Since limited specific  
17 information is available concerning THRs, this article aims at assessing reactivity in some  
18 THRs. To that end, the horses were exposed to two challenges and their behavioural and  
19 physiological responses were compared to jumping horses' (JH) reactions.

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## 1    **2.            Materials and methods**

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3            Eight horses were examined: five geldings and three female horses aged between three  
4 and nineteen years old. Four of them were therapeutic riding horses (TRH) and four were  
5 jumping horses (JH), used as controls. All the animals were housed in solid-sided boxstalls  
6 allowing them both visual and auditory contact and they all underwent a similar management  
7 pattern. The tests were carried out over eight days following a rest day in similar climate  
8 conditions. In every test day, two TRHs and two JHs underwent one test.

9            During the first test the horses were exposed to a restraint covering their head with a  
10 solid hood and they were tied by means of leads within their own boxes for one hour. In the  
11 second test a startle stimulus (a 40 cm long, red and white synthetic holiday garland shaken  
12 with a rustling noise inside the box) was introduced into each box where the animals were  
13 totally free to move. As soon as a horse approached the garland in order to explore it, the item  
14 was shaken by a person who was not visible to the animal. The test lasted 23 minutes.

15            The horses were continuously recorded as they underwent the behavioural tests. The  
16 video-recorder was placed on a tripod in front of each box door. The recorder's placement  
17 allowed observation of the whole body of each horse regardless of its position within the box.  
18 After starting recording, the operator left the box door in order to avoid any interference  
19 during the test. Video-recordings were analysed by means of the Observer Video-Pro  
20 software (Noldus et al., 2000). The following behaviours by the horses during the restraint  
21 test were considered: immobile, alert (ears stretched and leads strained, head and body still),  
22 head lowered, exploring the environment, pawing, head tossing, pulling back on the leads,  
23 moving. During the startle test, the following behaviours were examined: immobile, head  
24 lowered, looking at the garland, exploring the garland, moving, avoidance and flight attempts.

1 Heart rate (HR) and heart rate variability (HRV) were telemetrically recorded by  
2 means of two heart rate recorders Polar Vantage NV (POLAR ®) for each horse, during a  
3 basal period and during challenges. HRV describes variations of both instantaneous heart rate  
4 and interbeat intervals (Task Force of the ESC and the NASPE, 1996) and gives information  
5 about the sympathetic-parasympathetic autonomic balance. A preliminary visual analysis of  
6 data was carried out in order to identify artifacts caused by movements of electrodes on the  
7 skin or muscle contraction. For each horse, a five minute recording without artefacts was  
8 selected. The following time domain parameters were calculated (Marchant-Forde et al.,  
9 2004): average inter-beat interval (IBI), maximum and minimum R-R waves intervals  
10 (RRmax and Rrmin), standard deviation of the R-R intervals (SD) and the root mean square  
11 of successive differences (RMSSD). Frequency domain analysis was performed and the  
12 following parameters were calculated (Marchant-Forde et al., 2004): LF (0.01 – 0.07 Hz, Low  
13 Frequency, corresponding to the sympathetic nervous system activity), HF (0.07 – 0.5 Hz,  
14 High Frequency, corresponding to the parasympathetic autonomic nervous system activity)  
15 and LF/HF (corresponding to the modulation of the sympathetic versus vagal branches).

16 Before and immediately after every challenge, venous blood samples were taken at  
17 jugular level and put into silicone Vacutainer ® test tubes containing EDTA (1 mg/ml blood).  
18 Lymphocytes proliferation,  $\beta$ -endorphins and cortisol were determined (Sacerdote et al.,  
19 1994).

20 Hematochemical and immune parameters were analysed by non-parametric Wilcoxon  
21 Match Paired Test (SPSS, 2003). Integrated behaviour and heart rate analysis were carried  
22 out. For each behaviour average durations and relative heart rates ( $\pm$ SD) were calculated. The  
23 relevant data were analysed by means of the non-parametric analysis of variance. Heart rate  
24 data were analysed by means of the non-parametric variance analysis and General Linear  
25 Model analysis (GLM) for repeated measures (SPSS, 2003). Heart rate variability was

1 calculated by means of the index method and the frequency method and the relevant data were  
2 analysed by Wilcoxon Match Paired test.

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### 5 **3. Results**

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7 The horses undergoing the tests did not show any aggressive behaviour and/or did not  
8 make any significant attempt to run off. During the challenges, both TRH and JH spent more  
9 time standing still than performing other behaviours (ANOVA:  $F_{1,7}=6.12$   $P<0.05$ ). Table 1  
10 features time periods associated with behaviours. Altogether the two groups showed different  
11 values in terms of heart rate, which was lower in JHs (ANOVA:  $F_{1,1}=9.25$   $P<0.05$ ). No  
12 statistically significant difference between behaviours during the startle test was found by  
13 analysing the relevant heart rates. Nevertheless, THRs adopting the "look at the garland"  
14 behaviour showed higher average HR values (55,5 bpm) with respect to other behaviours  
15 (31,8 bpm). During the first minute after shaking the garland TRHs had a larger increase in  
16 HR (135 bpm, WILCOXON:  $S=26$   $Z=2,17$ ;  $p<0.05$ ) than JHs (64 bpm, WILCOXON:  $S=26$   
17  $Z=2,17$ ;  $p<0.05$ ). During the restraint test, HR values varied according to the behaviour  
18 (ANOVA:  $F_{1,7}=3.32$   $P<0.05$ ). Figure 1 points out that the highest HR values were recorded  
19 in both groups while horses were "pawing" (GLM:  $F_{1,7}=3.32$   $P<0.005$ ). Figure 2 features the  
20 HR evolution in both groups during the restraint. Time has a statistically significant impact on  
21 HR (GLM:  $F_{1,11}=8,73$   $P=0.0001$ ) despite similar patterns in both groups. Table 2 features  
22 the HRV average values. The table reports the comparisons made between the basal period  
23 and the recordings after the restraint and the startle test on both groups (TRHs and JHs). No  
24 significant difference was found in any of the parameters. Not even the LF/HF parameter,  
25 which is a well-known indicator of the modulation between the sympathetic and

1 parasympathetic autonomic nervous system, showed a clear trend. Lymphocyte proliferation  
2 decreased significantly after removing the hood in both TRHs and JHs (WILCOXON:  $S=88$   
3  $Z=2.05$   $P<0.05$ ). Cortisol basal values showed no statistically significant differences between  
4 TRHs and JHs ( $5\mu\text{g}/100\text{ml}$  in TRHs and  $4\mu\text{g}/100\text{ml}$  in JHs). Nevertheless, after the restraint  
5 JHs showed a trend towards increasing concentration ( $P = 0,06$ ). As for the other  
6 hematochemical parameters, no significant difference was determined between basal values  
7 and test values.

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#### 9 **4. Discussion**

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11 This study focused on various parameters since literature relating to stress indicators  
12 reliability in horses is quite fragmentary (Baucus et al., 1990; Colborn et al., 1991; Rugh et  
13 al., 1992; McCarthy et al., 1993; Canali et al., 1996; Thayer et al., 1997; Wolff et al., 1997;  
14 McGreevy and Nicol, 1998; Anderson et al., 1999; Minero et al., 1999; Vierin et al., 1999).  
15 Hence it was considered important to compare and combine various physiological and  
16 behavioural data. As for the behaviours recorded during the test, no excessive reactivity by  
17 the horses was shown. Should TRHs have been too reactive, their suitability as therapeutic  
18 animals should have been questioned. The horses were watchful and responsive to external  
19 stimuli. During the restraint they were "alert" for long periods of time. The HR difference  
20 shown by the two groups could be related to the effects of a more intensive training of the JHs  
21 with respect to TRHs, which are basically used only for therapeutic purposes. The highest HR  
22 values were recorded during the restraint test while the horses were "pawing". This behaviour  
23 is often seen as a reaction to frustration, a displacement activity (Ödberg, 1973). Horses  
24 normally behave this way while waiting for feed, as an attempt to get close to another animal,  
25 or to rouse a foal in prone position (Haupt, 2000). In this study, "pawing" can easily be

1 interpreted as a response to restraint since the horses were tied. As for the fear reactivity test,  
2 the highest average HR was recorded during the "look at the garland" behaviour. This was  
3 probably due to an increase in the sympathetic nervous system activity, although the horses  
4 were not significantly aggressive and/or did not try to run off. A decrease in the lymphocytes  
5 blastogenic reaction to mitogens was observed during the restraint test in both TRHs and JHs.  
6 This might be due to the fact that the horses had to face a stressor they could not avoid.  
7 Literature (Laudenslager and Ryan, 1983) shows that in these cases lymphocyte proliferation  
8 decreases or disappears, while it actually increases when an animal is given the opportunity to  
9 react by adapting to the new situation (Laudenslager et al., 1983). The individual differences  
10 shown by the horses as a reaction to these challenges might be associated with different  
11 adaptation skills and temperaments (Visser et al., 2003) regardless of their ordinary activities.  
12 No statistically significant data were found in some parameters such as heart rate variability  
13 or cortisol. This might be due to high individual variability and to the limited number of  
14 horses being examined. This conclusion needs further investigation with a broader sample.

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## 16 **5. Conclusion**

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18 The horses reacted differently to the two challenges. During the startle test, differences  
19 were recorded between basal values and test values only for the most straightforward and  
20 sensitive parameters (behaviour and heart rate), while the restraint caused a stress reaction  
21 implying a behavioural and physiological response at various levels. It is possible to assume  
22 that TRHs are familiar with everyday stimuli associated with their specific activity. However  
23 they react to novel stressors as stated by Anderson et al. (1999). As a consequence, they might  
24 react to unfamiliar stimuli despite the specific training they should undergo and the routine  
25 work they perform. Such studies are difficult to plan as they demand a sufficient number of



1 TRHs with similar management patterns (which is not common) and active involvement and  
2 dedicated cooperation by the Center staff. However they are useful since limited information  
3 is available concerning horses' adaptation skills to this kind of work and since information is  
4 important in order to plan therapeutic activities and ensure their effectiveness.

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