Ann Ist Super Sanità 2015 | Vol. 51, No. 1: 40-51
DOI: 10.4415/ANN_15_01_08

ORIGINAL ARTICLES AND REVIEWS

The role of associative and non-associative learning in the training of horses and implications for the welfare (a review)

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Abstract
Horses were domesticated 6000 years ago and since then different types of approaches have been developed to enhance the horse's wellbeing and the human-horse relationship. Even though horse training is an increasingly important research area and many articles have been published on the subject, equitation is still the sport with the highest rate of human injuries, and a significant percentage of horses are sold or slaughtered due to behavioral problems. One explanation for this data is that the human-horse relationship is complex and the communication between humans and horses has not yet been accurately developed. Thus, this review addresses correct horse training based on scientific knowledge in animal learning and psychology. Specifically, it starts from the basic communication between humans and horses and then focuses on associative and non-associative learning, with many practical outcomes in horse management from the ground and under saddle. Finally, it highlights the common mistakes in the use of negative reinforcement, as well as all the implications that improper training could have on horse welfare. Increased levels of competence in horse training could be useful for equine technicians, owners, breeders, veterinarians, and scientists, in order to safeguard horse welfare, and also to reduce the number of human injuries and economic loss for civil society and the public health system.

Key words
- animal learning
- training
- horse
- welfare

INTRODUCTION

In recent decades, the horse has been used in a variety of roles that is unparalleled to other species. Horses are considered workforce and a source of meat in developing countries, whereas in Western society the horse has become a companion animal used for leisure and sports, developing an industry estimated at tens of billions of euros in Europe alone [1]. In addition, the horse has recently played an important role in Animal-Assisted Activities and Therapies [2, 3].

There is a widespread notion regarding horse and human interaction that the horse should be submissive and docile to humans [4]. It is believed that during the introductory training process, the domination by a person and the subjugation of the horse are essential for success [5]. Taming techniques have two common elements: isolation of the horse from its peers, and ensuring that the horse accepts its attempts of avoidance and escape will be unsuccessful [6]. These elements are present even without apparent use of coercion, as in "natural horsemanship" (codified techniques of horse training, developed in the USA, and known as "round pen techniques" from the name of the circular enclosure where they are generally implemented) that seem, to the layman's eye, more ethically acceptable [7, 8].

Even though horses have been domesticated for more than 6000 years and different types of taming have been developed, horseback riding is the sport/leisure activity with the highest rate of human injuries [9-11]. Furthermore, many horses are "lost" due to the onset of behavioral problems that make them unfit to perform the task for which they are bred [12, 13]. Consequently, a high percentage of horses are slaughtered at a young age or before reaching normal retirement age [14]. In

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the USA, the percentage of horses slaughtered for behavioral problems is about 10% [15]. Consequently, there is a significant loss of economic resources for civil society and the equestrian industry; this is also indicative of compromised conditions of physical and mental well-being for horses [16]. Very recently it has been demonstrated that horses in these types of conditions can show depressive-like behaviors [17] and learned helplessness could be a consequence of several years of improper management [18]. For instance, older horses used for activities that require calm and non-reactive animals (*i.e.* riding schools for beginners or equine assisted activities and therapy) may experience negative mood states, although they appear calm and show little behavioral reactivity. All of these issues suggest that the developed communication between human and horse is not yet correctly refined [9].

Furthermore, while the influence of emotional and cognitive performance is widely studied in the human athlete [19], demonstrating that mental health and well-being are essential to good performance [20], this field is still lacking research in horses. Within equitation activities (sports or leisure or animal-assisted activity and therapy), two biologically and evolutionarily different living beings (horse and human) need to collaborate and communicate to achieve positive results [21]. Competitive horseback riding is the only Olympic sport with this unique requirement. It becomes evident that in order to obtain good results within the horse-human relationship, it is essential to consider the psychology of the horse and its emotional involvement. Riders and trainers should carefully take into account all factors affecting the welfare of their horses [22], especially in the early stages of training when psychological rules of animal learning are critical to develop a balanced relationship with humans for both riding and handling purposes.

This article critically reviews the most recent published papers to provide a comprehensive, detailed, and deeper understanding on how associative and non-associative learning functions when working with horses. Despite the fact that some review articles have been already published on this topic, this review highlights some examples of practical outcomes in training from the ground and under saddle. This was done to underline the application of learning theory in practice; this review may become a useful tool in horse training and management. The non-associative and associative learning rules (see *Table 1* for the most relevant definitions) should be deeply understood by all individuals working with horses, not only to improve the physical and psychological welfare of horses, but also to reduce accidents and the number of horses lost to behavioral problems caused by an improper relationship with a person.

The key to resolving these issues and improving the welfare of horses is recognizing the lack of the use of scientific knowledge in the horse-human relationship, regardless of the activity in which they are involved (sports, leisure, assisted activity and therapy, etc.) [22].

In recent years, studies on equine behavior have made significant progress. Horses recognize each other individually [23] and recognize humans [24]. They possess sophisticated cognitive strategies to maintain social equilibrium [25] and have a long [26] and short term memory capacity [27]. Furthermore, recently patterns of intraspecific communication have been highlighted in horses [28]. Therefore, the complexity of the horse's mind and behavior indicates that learning skills of horses are qualitatively the same as those of other domestic mammals. Once professional and leisure riders are able to properly apply the rules of animal learning to horse training and handling, their scientific knowledge should also be expanded to these more complex cognitive aspects related to horse-human relationships and stable environments, not included in the topics of this review.

This article starts by outlining proper communication with horses through the use of pressure, then presents how rules of learning theory should be applied to the training of horses, explaining how positive and negative reinforcements work. Practical procedures and outcomes are then suggested for handling horses from the ground and under saddle. The review then addresses how classical conditioning may help to reduce stimuli administered to the horse. Finally, the authors discuss common mistakes in equine training, such as the risk of habituation to pressure stimuli and the dangerous boundary between the correct application of negative reinforcement and punishment.

COMMUNICATION WITH THE HORSE THROUGH PRESSURE

Practical experience tells us that horses often exhibit unexpected and defensive behavior without a plausible explanation. There is the possibility that these behaviors are simply due to communicative signals performed by the human, signals that humans are often unaware of giving to the horse.

The actions performed by humans with the legs on the horse's side, hands on the reins, and body position in the saddle affect the ridden behavior of the horse [29]. Human actions also affect the horse when led from the ground with a halter or during handling for medication or clinical investigations. During these activities, most gestures of the person are physical actions transmitted to the horse, thus representing a form of physical communication. Every time a rein is pulled, the whip is used, the horse is touched with the rider's leg or led by hand with the halter, the mechanical action performed with the hands or other parts of the human body is transmitted to the horse's body [30-32]. Essentially, physical forces are applied to induce behavioral responses in horses and become "pressures," stimuli on the body of the horse, causing change in its behavior [33]. For example, simply inserting a finger between the horse's skin and the various instruments (*i.e.* bit, noseband, bridle, saddle, twitch, curb chain, bit-less…) is a useful and easy exercise to understand the pressure exerted on the horse's body by every action performed by humans [34]. These pressures can be measured in some cases [30, 35] and are often amplified by the use of martingales, draw reins and other training devices [36]. The pressure applied on the body of the horse (*Figure 1*) is an aversive stimulus, which creates discomfort [37]. This discomfort

Table 1
Definitions of animal's learning. The psychological rules of learning are the same in horses and other animals as well

Relationship	The emerging bond from a series of interactions that partners have. It is based on past experiences and expectations of the other individual's responses [78].
Communication	The activity of conveying information through the exchange of thoughts, messages, or feelings, as by vocal and visuals signals, or behavior. It is the meaningful exchange of information between two or more living creatures [79].
Stimulus	Any appreciable change in the environment that causes a behavioral response in the animal [70].
Learning	The information obtained from the interaction between an environmental stimulus and the elicited behavior will form the experience, according to which the animal will change its behavior in the presence of that stimulus when it will reoccur in the future [70]. Broadly, animals learn to use the information coming from the environment to change their behavior in the most advantageous manner to them [73].
Training	Training suppresses undesirable behavior and enhances desirable natural or new behavioral responses by punishing or reinforcing them with the deliberate or accidental application of learning theory [68]. The goal of training is to lead the animal to perform a predictable behavior as a result of the appearance of specific signals-behaviors that should be resistant to extinction [80].
Associative learning	Process that allows the animal to establish the connection between two events in a relationship of reciprocity between them [70].
Non-associative learning	Refers to a relatively permanent change in the strength of a behavioral response to a single stimulus due to repeated exposure to that stimulus [70].
Operant conditioning	Is a type of associative learning in which an individual's voluntary behavior is modified by its antecedents and consequences [81]. It works by giving or taking away rewards or punishments (discomforts) when the horse performs a desired behavior through the chain: stimulus - response - reinforcement [68]. In operant conditioning it is the animal's behavior that determines the progression of the reinforcement. Therefore, it allows the animal to associate two events over which it has control [70].
Classical conditioning	Is a type of associative learning whereby behavioral response becomes elicited from a conditioned stimulus [82]. With classical conditioning, animals learn which environmental cues predict future events so that they can behave accordingly [68]. In such cases, the animal has no control over events; and the response is not under the control of the animal. Classical conditioning increases the predictability of environmental stimuli [70].
Reinforcement	Any event that increases the frequency of a certain behavior and makes it more likely to of occur in the future [70]. The reinforcement needs to be something biologically relevant for the animal (the removal of discomfort or the appearance of food), so it is highly motivated to obtain it. • Negative reinforcement is the subtraction of something aversive [83]. • Positive reinforcement is the addiction of something pleasant [81].
Primary and secondary reinforcements	Primary reinforcements are any resources that animals have evolved to seek (food, water, sex, play, freedom, companionship), whereas secondary reinforcement are stimuli which are not intrinsically rewarding but that can be associated with primary reinforcement (through classical conditioning) [29].
Continuous reinforcement	Each correct behavior of the animal is reinforced [68].
Intermittent reinforcement	It reinforces a behavioral response followed by the non-reinforcement of a certain number of behavioral responses [68]. Intermittent reinforcement is divided into: • intermittent reinforcement fixed ratio (when the reinforcement is repeated with the same frequency alternating one response reinforced with a constant number of not reinforced responses); • intermittent reinforcement variable ratio (when the reinforcement is administered with random mode compared to the number of correct answers).
Punishment	Punishment is any action that makes the occurrence of a behavior less likely to be performed in the future [29]. • Positive punishment is to add something undesirable or painful; • negative punishment is to remove of something desirable by the animals.
Shaping	Training performed by putting together the elements of the behavior, starting from the horse's natural responses and ending, through subsequent approximation, to a complex pattern of an essentially unnatural behavior [68].

can be of varying intensity, from very light and almost imperceptible at the sensory level, to very strong and eventually painful if legs and hands are used improperly [31, 38]. The pressure (hence the discomfort) is then removed when the horse performs the desired behavior. The horse stops when the rider pulls the rein because this behavior helps it to reduce the pressure on the muzzle, nose, or mouth [33]. Therefore, during training, horses learn that pressure applied by the rider through the reins disappears when they stop or slow down the pace, the pressure of the rider's legs disappears when they move forward, and so on.

THE TRAINING OF HORSES

To be effective and ethologically correct, training should include the application of pressure and its immediate removal when the horse behaves as requested [22]. When this does not happen and the release of pressure (the removal of discomfort) is performed incorrectly, horses may develop conflict and defensive behaviors, making it difficult to manage them. This mainly occurs when pressure becomes pain [39, 40]. These distinctions are particularly important in the early stages of training. Despite the large number of sports and recreational activities in which horses are involved,

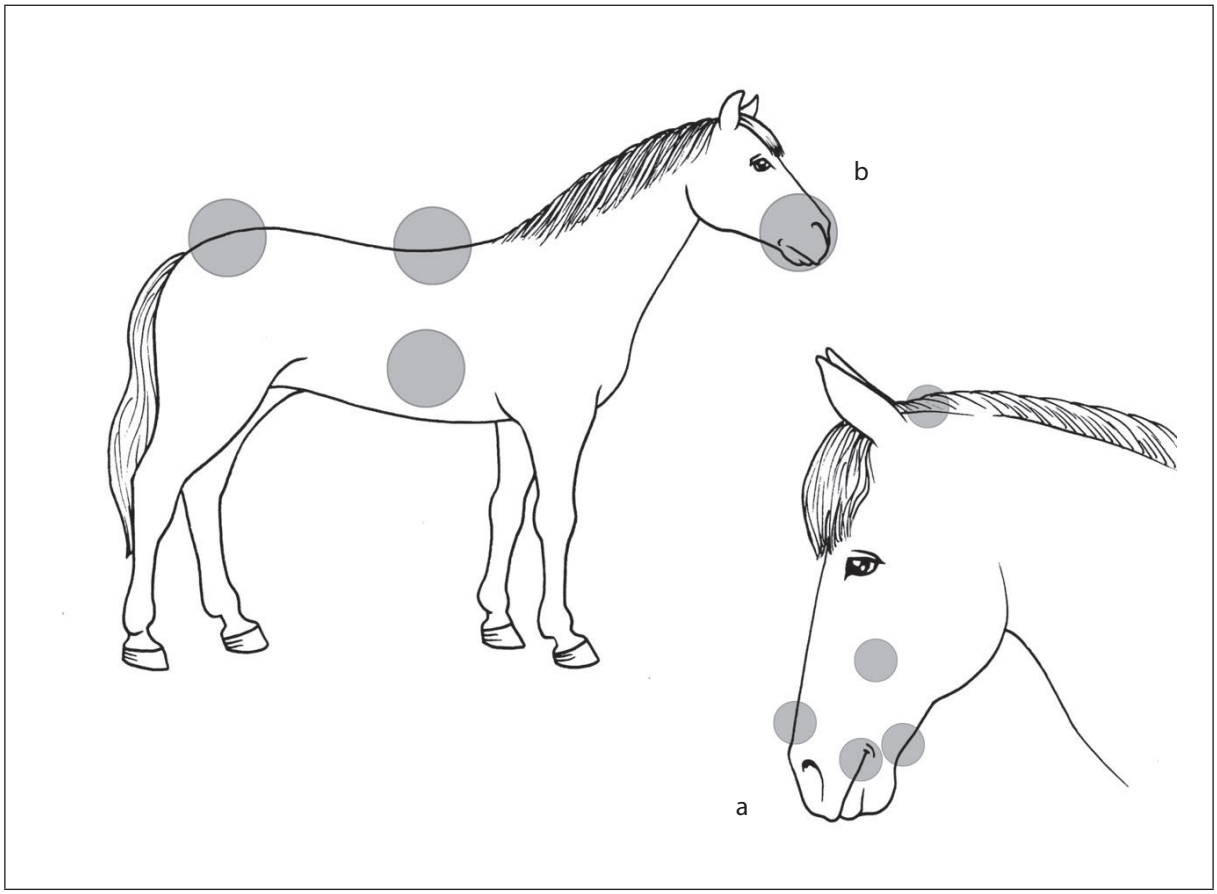

Figure 1
The main points of the horse's body (gray areas) where pressure is exerted both under saddle and when handling from the ground. On the horse's head (a), the points of pressure vary according to the type of bridle used (with or without bit). On the contrary, the pressure places on the horse's body are more homogeneous (b), and are mainly due to the presence of the rider while working under the saddle.

there are four basic responses on which the training of horses is built: forward, stop, turn on the forehand, and turn on the haunches [41]. At the beginning of the training, which is the critical phase of the proper relationship between horse and human, horses learn these basic responses through negative reinforcement [42]. Most actions that take place in handling a horse from the ground and from the saddle are related to the use of negative reinforcement [37]. Essentially, the training is based on the capabilities of horses to avoid discomfort [43] and negative reinforcement can also be used without touching the horse. An example of this is the use of fear as an aversive stimulus, which results in the horse galloping around the perimeter of the round pen [6].

The main purpose of training is to provide signals that the horse learns to associate with a specific behavior. When this does not happen and the horse performs unexpected or unwanted behaviors, this result means that the training is not adequate in establishing the association between the desired signal (the pressure) and the desired behavior, and the reinforcement used to create this association is insufficient in overcoming the horse's motivation to perform other behaviors [22]. Therefore, unexpected and defensive behavior may be the result of communication signals that are unclear to the horse [5]. Even so, in the face of many behavioral responses required by training, the points of the horse's body where riders can apply pressure are relatively few [41]. That is, during the training process the rider has at his disposal a limited "vocabulary" for asking the horse a considerable number of behavioral responses. As discussed, the communication between human and horse from the ground and from the saddle takes place through the application of pressure, administered with the aid (either natural: such as hands, legs or seat, or artificial: spurs, whip and other training devices). This communication is actually a binary language of "*Yes, this is what I want*" (release of pressure) and "*No, this is not what I want*" (application of pressure). The question is how can we effectively communicate with the horse using only Yes and No? Take, for example, the game of finding a hidden object following the "hot" (Yes) or "cold" (No) clues, depending on whether the person playing moves closer to the goal. The secret to finding the object lies in the skill of the person who is driving the game by saying "hot" (Yes) as soon as the subject makes the slightest movement in the correct direction and then continues to be more and more accurate. If the person who is driving the game is not ready to say "hot" (Yes) at the slightest hint of movement in the correct direction, the sub-

ject will move based on chance. This lack of clarity with orders will lead to confusion and frustration, as most likely happens for horses [43]. Therefore, the key to horses' welfare is to understand how the reinforcement works, especially the timing when the pressure applied by the rider or caretaker must be released (or the food provided) during training and handling management.

Positive and negative reinforcement in the training of horses

There are two main types of reinforcement: positive and negative (*Table 1*). Reinforcement, by definition, is always advantageous, because it has a value for the animal. Depending on the context of the training and its goals, positive and negative reinforcement can be easily implemented from the ground, whereas when working from the saddle, riders cannot adequately apply positive reinforcement using food [6, 45]. Despite these issues, positive reinforcement should be considered while handling the horse from the ground and a mixed strategy (using both positive and negative reinforcements) could present a balanced solution for considering the horse's welfare [46].

The use of aversive stimuli has been common practice when teaching horses to load on a trailer or correcting loading issues. Some halters and head-collars, for example, are designed to exert pressure at various points on the head of the horse when it pulls away from the caretaker [47]. Further practical evidence suggests wrapping a long rope around the hindquarters of a non-collaborative horse when attempting to load it, or prodding the horse with the bristles of broom, in order to pressure it from behind. Subsequent correct responses of approach to the loading ramp are therefore negatively reinforced as they result in the termination of these pressures.

Negatively reinforcing forward motion by removing an aversive stimulus (pressure) is effective only if it can be performed consistently and with correct timing. However, a number of other potentially dangerous avoidance behaviors (such as rearing or bolting) that do not result in loading could also lead to the termination of pressure for the horse [47]. The risk is that an inexperienced trainer could negatively reinforce incorrect or dangerous behaviors, thus cultivating subsequent loading problems. Furthermore, increasing aversive stimuli is likely to induce habituation, which leads to a reduction in response [48]. This would render strategies employed ineffective and potentially lead to abuse of the animal [49]. Overall, the aversive treatment of problematic behavior (through negative reinforcement), such as trailer loading, could lead to further undesirable behaviors and could be inadequate in overcoming common problems [47]. Animals trained through positive reinforcement seem to be easier to load and appear less stressed [50]. Thus, it is essential that aversive (*i.e.* negative reinforcement) and non-aversive (*i.e.* positive reinforcement) training methods are balanced in order to optimize learning abilities of horses and improve their welfare [51] and to evaluate learning performance people should take into account visual attention of horses [52].

In a comparative study between positive and negative reinforcement training, horses trained with positive reinforcement were more likely to display the target behavior and show increased motivation to participate in training activities [46]. Therefore, applying positive reinforcement during common handling procedures could lead horses to better cope with training-induced stress [51], mainly from the ground. Horses can be trained to perform various tests to obtain a food-based reinforcement, such as pressing a lever [53], completing a maze [54], and choosing between containers of different colors [55, 56] or forms [57]. The same principles can be applied to training a horse to load on a trailer by means of positive reinforcement [47].

Handling the horse from the ground (with practical outcomes)

When a horse performs an undesirable behavior, instead of attributing anthropomorphic adjectives (evil, vindictive, submissive, dominant, etc.) to it, we should remember that horses tend to repeat any behavior that precedes a reward such as food, or relief from discomfort with greater intensity and frequency [33]. Therefore, firstly the reinforcements (often unobvious) that led the horse to behave that way should be determined. With the application of a sudden pressure, the horse can be induced to display a defensive or escape behavior to stop it. The more unexpected and severe the pressure is, the greater the possibility that the horse will perform one of these behaviors [6, 58].

Most of the actions by the veterinarian put pressure on the horse, often of high intensity. For example, an injection is a sudden and high intensity pressure, exceeding the threshold of pain. The insertion of the needle (*stimulus*), especially initially, can induce the horse to perform defensive behaviors, which may be mild (moving the muscles of the neck, shaking the head, etc.) or strong (rearing, etc.) (*response*). If one of these behaviors during the injection causes the needle to slip off the skin, the horse learns that this behavior functions to remove the discomfort created by the needle (*reinforcement*), so the possibility that this behavior will be repeated by the horse at the next injection becomes high (*Figure 2*). By applying the rules of associative learning, we can train the horse to perform a more functional behavior, rather than a defensive behavior (dangerous for both horse and human) [22]. In the case of injections, this would be the behavior of standing still. If the horse reacts to the injection, instead of immediately inserting the needle, the handler should tap the horse's neck continuously with a finger, simulating the syringe and continuing this pressure as soon as the horse's first movement occurs. As a result the horse will not associate his movement with the prevention of the injection. As soon as the horse stands still, at least for a moment, the handler should stop tapping the finger on its neck. With a sufficient number of repetitions of this sequence, the horse will learn that the behavior which stops the simulated pressure of the syringe is standing still (*Figure 2*). Another example involves horses that shake their heads, moving and trying to rear during clipping. In this case it is non-associative

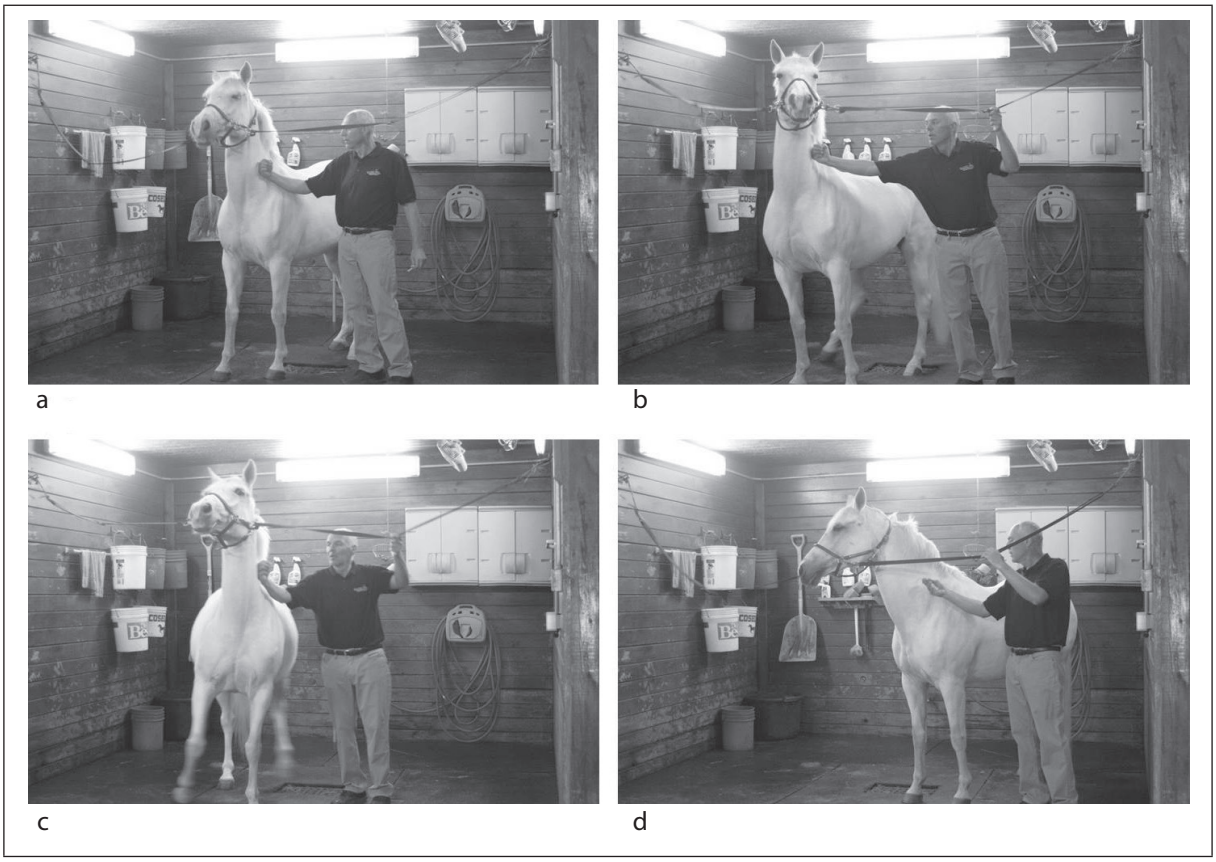

Figure 2
The insertion of the needle can cause the horse to display defensive behaviors. If these behaviors are functional to stop the action of the needle, the horse learns to repeat that behavior at the next injection. To avoid such situations, instead of immediately inserting the needle into the skin, we can tap the horse's neck with a finger simulating the syringe (a). In this way, we can continue the pressure during the execution of defensive behaviors (b). The horse therefore will not associate its movements to the cessation of injection (c). As soon as the horse stops, even only for a moment, we stop the tap of the finger on its neck. The horse learns that the behavior that stops the simulated pressure of the syringe is standing still (d) and is more likely to remain stationary during the injection.

learning (habituation) and negative reinforcement that regulate the behavioral response [59]. Through habituation, in most cases horses learn that the passing of the clipper on the body is not an unpleasant stimulus. For this to happen, it is important that the horse stands still while clipping; but if it shakes, moves to the side, or performs other aversive behaviors, and the clippers are stopped in that moment, the horse will learn that there are behaviors that it can perform to stop the clipping. In other words, when the clipping begins and the horse moves, the handler should continue the clipping without interruption. When none of the horse's aversive behaviors cause the clipping to stop, the horse becomes habituated to the clippers (which, contrary to the syringe, usually does not induce pain) and thus, stops reacting. This sequence refers to the application of non-associative learning and works well with horses clipped for the first time, especially when clipping the thorax and abdomen, because even if the horse moves, generally the contact with its body can be maintained. The process becomes more complicated when the clipping gets closer to the head or legs. While clipping these areas (*stimulus*), the horse is able to get away with a sudden movement (*response*) and stop the action of the clipper (*reinforcement*). If this occurs, even with the continuation of the work of desensitization, it will be difficult to solve the problem because, through operant conditioning, the horse has just learned a behavior to avoid the action of the clipper. To prevent the occurrence of this problem, it is necessary to employ negative reinforcement, releasing the pressure (*i.e.*, by interrupting the action of the clipper) at the proper time. The handler should turn on the clippers and get closer to the horse. As soon as the horse begins to show defensive behaviors, even minor ones, the handler should stop and maintain the position with the clippers switched on. When the horse stands still, the handler should immediately turn off the clippers. This sequence needs to be repeated several times until the horse associates the position (standing still) with the action of turning the clippers off. It is important to turn off the clippers often when the horse is still to reinforce its behavior and clip intermittently, rather than continuously as when the horse is already accustomed to standing still. However, once the association has taken place, the handler should simply turn off the clippers at

intermittent periods during clipping so the horse will stand still for a longer time while the clippers are running, because it has learned that eventually the clippers will stop (according to the rules of variable schedule of reinforcement). It is important to remember that correct application of the rules of animal learning requires concentration on what the horse is doing and what the handler is doing. To correctly reinforce a behavior, the handler must intervene in a span of a very few seconds (the closer the reinforcement, *i.e.* the removal of the pressure, is to the behavior, the greater the possibility of creating the association) immediately after the horse performed the correct behavior [60].

Rules of animal learning can also be useful in the stable for horses that kick stall doors, bite bars, and/or attack people and neighbors when approaching feeding time. Keeping a horse in the stable alters its natural behavior [61, 62]. In the stall, the horse stands still for most of the day, often without social contact, and eats for a shorter period than when out on pasture. This type of management can lead to the onset of destructive and abnormal behaviors [63], which offset natural behaviors such as walking and chewing and, with the passage of time, can develop into fixed behaviors, becoming compulsive and then stereotyped [62]. In natural life there are different patterns of behavior related to foraging (scratching with the foreleg, threatening herd members, etc.) that the horse tends to repeat in the stall, where the situation is exacerbated by the fact that food is available only for short time periods during the day [62]. This may lead to the occurrence of these behaviors (scratching, kicking, biting, etc.) at every mealtime. It may happen that the caretaker administering the food does not pay attention to the horse's behavior. In these cases, operant conditioning teaches us that if we put the food into the feed bucket when the horse is performing a destructive behavior (with perfect, but unintended, timing of reinforcement), that behavior is then reinforced. For example, if we put the food in when the horse is kicking, we are teaching it that a kick is rewarded with food. With time, this association will become fixed and the horse will use more and more of this behavior as the meal approaches. The unaware action of the caretaker (who neglects to notice behavior of the horse while feeding it) means that, once learned, these behaviors are reinforced with an intermittent reinforcement variable ratio, which is one of the strongest in maintaining fixed behavior because it introduces a certain degree of unpredictability in the arrival of the reinforcement [29]. This is similar to the case of the human being who plays slot machines, where an award intermittently and by chance from the behavior of pulling the lever is sufficient to induce people to repeat this movement regularly [64].

The best strategy to preventing the horse from learning these types of behaviors is a knowledge base of associative learning rules and avoiding unconsciously reinforcing them. However, if these undesired behaviors are unconsciously reinforced, the intermittent positive reinforcement of other behavior through operant conditioning techniques allows us to teach new behaviors that can replace the unwanted ones [65].

Working under saddle (with practical outcomes)

The rules of learning are the same with mounted horses. For example, we can analyze how to teach the horse to leg yield. The rider applies the pressure of the inner leg while resisting with reins. At this point the horse will probably begin to move forward, as this is the previously learned behavior associated with the pressure of the leg. This time, instead of releasing the pressure, the rider will maintain it (*No, this is not what I want*) until the horse will show even minimal lateral movement. At that time the rider will immediately release the pressure (*Yes, this is what I want*). The rider will then have properly reinforced the slightest lateral movement of the horse through the chain: stimulus (pressure) – response (lateral movement) – reinforcement (immediate release of pressure). What errors may be committed? It is possible that the first time the pressure is applied, the horse will perform other behaviors (shakings its head, trying to bite the rider's leg, or, in extreme cases bucking or rearing). The horse may perform these behaviors to stop the pressure from the reins and legs before the desired behavior (to perform a lateral step) [35]. If one of these incorrect behaviors results in removal of the pressure, the horse will learn that bucking or rearing relieves it from the pressure (*Figure 3*). There are other errors the rider can commit. If the horse is believed to be performing the leg yield because the pressure of the inner leg moves it to the side, the rider will continue to press while it's moving. In this way the rider is maintaining the pressure while the horse performs the desired behavior. Therefore, despite the horse giving the correct answer, the rider is saying to it, "*No, this is not what I want*." This will probably create confusion to the horse, pushing it to try other behaviors to gain relief from the pressure. These defensive behaviors will be shown with greater frequency and intensity when the same pressures arise again in the future (for an overview about behavior of mounted horses see [32, 35, 66, 67]). It is essential to underline that horses are unlikely to be able to assimilate all the components of a complex behavior at once. Therefore, the training process requires putting together a step-by-step sequence comprised of single elements of the final complex behavior required, with successive approximations until the final result is achieved, following the shaping procedure [68].

THE POWER OF CLASSICAL CONDITIONING AND ITS IMPLICATION FOR THE WELFARE OF HORSES

Through classical conditioning, a stimulus that originally has no meaning for the animal is associated with an unconditioned stimulus that can be used as a reinforcement (or punishment). This means that once a behavior has been learned by negative reinforcement, the horse will use other stimuli to predict the arrival of the reinforcement, *i.e.* the removal of discomfort. Riders use classical conditioning when replacing pressure stimuli used in the initial stages of training with new stimuli that were initially neutral stimuli (they evoked no response) [49]. This is called secondary reinforcement-stimuli which can be words, gestures, or other

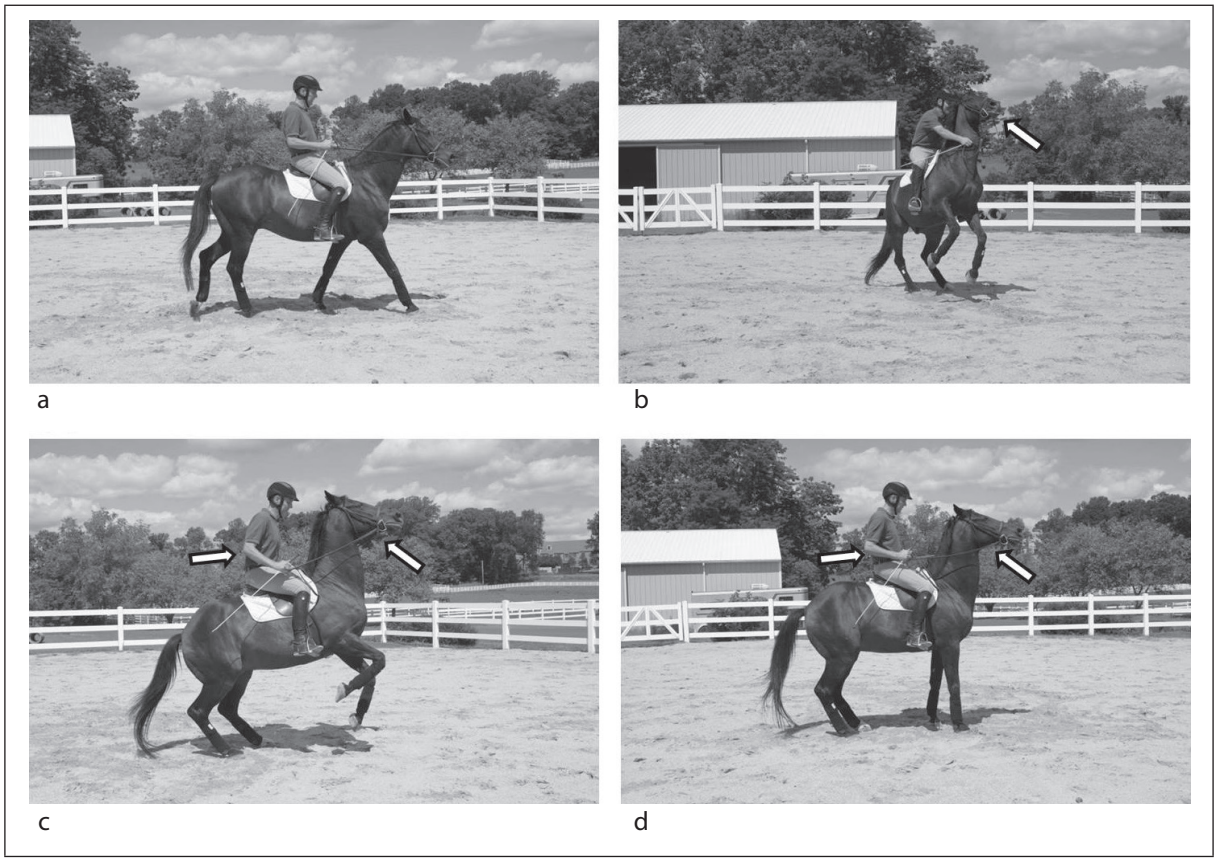

Figure 3
The onset of defensive behaviors, such as rearing, is often the result of errors in the removal of the pressure. With the appearance of the pressure from the rider's legs and reins (a) the horse could move forward, or display defensive behaviors to remove that pressure. For example, if the horse rears and the rider, to remain in balance, embraces the horse with the hand and releases the pressure (arrow, b), the horse learns that rearing does stop rein pressure and tends to repeat this behavior when the same pressure occurs in the future. To prevent the horse from learning to rear, it is essential to keep the pressure on the reins during rearing (arrows, c and d) until the horse terminates such behavior. The pressures are then immediately released when the horse performs the desired behavior, such as walking forward.

pressure, such as the use of seat position [37]. This could have welfare implications for horses because the classical conditioning permits the rider to substitute the adverse stimuli of the negative reinforcement sequence with the use of light pressure or other non-invasive stimuli [68].

Due to classical conditioning associations, a horse can also show defensive or escape behaviors in an unexpected way without seemingly plausible reason [68]. When defensive behaviors have been learned, the horse can, through classical conditioning, predict the "injection" event because it will have associated a series of stimuli (a certain person who enters the stall, the smell, the sight of the syringe, etc.) that predicts the injection and it will show defensive behavior in advance. The same can happen when working under saddle. If a horse has learned to rear because of errors in the removal of the pressure by the rider during the application of negative reinforcement, it will associate other stimuli as prediction of the pressure's appearance through classical conditioning (even the simple entry into a competition or training arena) [22], and it will display the behavior of "rearing" before the onset of pressure that caused it has been reached. The lack of knowledge of these rules could be a source of behavior problems, because the stimulus that the horse chooses to predict the arrival of a pressure that has been already associated with defensive behaviors may be an unrecognizable stimulus, and therefore beyond our control. These defensive behaviors without notice, in advance and without apparent reason, are also called "anticipatory behavior" [69].

Some variants of classical conditioning are "blocking" and "overshadowing". They indicate how the simple coupling of a stimulus with reinforcement is not sufficient to produce the necessary associative learning, but rather that the animal makes a choice regarding which stimulus best predicts the arrival of the reinforcement [70]. These phenomena are very important for horses' training, both while working under saddle and from the ground [49, 71]. The more stimuli (pressures) that are concurrently given by the rider, the greater the chance is that the horse performs its selection, regardless of the will of the rider, about the stimulus which best predicts the removal of the pressure and implements behaviors that are unexpected to the rider [71]. An example can be seen in the canter departure that is requested with

light pressure from the leg aids [59]. While applying pressure with legs, the rider usually perform a thrusting movement with the pelvis on the saddle that also produces light pressure. By the blocking/overshadowing (in this case it is difficult to say which of the two singularities is actually involved), the horse could choose to respond to the pelvic movement and fail to pick up the canter from the leg aids. As a result the rider, trying to reestablish the correct negative reinforcement sequence to obtain the canter departure, may unknowingly think that the horse is unresponsive to the leg aid and consequently increases the leg pressure with the use of spurs or a whip, often exceeding the horse's threshold of pain. Similar situations to this one may lead to training the horse in a frequent state of pain, causing welfare concerns.

THE RISK OF THE HABITUATION

Non-associative learning is widely used in training to ensure a decrease in the frequency and intensity of undesirable behaviors performed by the horse, such as escape and defensive behaviors, and to habituate to new objects and environments [22, 72]. In horses, a common response to unfamiliar situations is to put distance between themselves and potential dangers by avoidance behavior [73]. With repeated exposure, horses habituate to their surroundings and cease to avoid nonthreatening stimuli [68]. For example, a naive horse may shy away when touched by a novel stimulus (such as a saddle pad or splint boots), but the response will diminish with repeated exposure [59]. In order to effectively train a horse with habituation, the stimulus offered must be equal and constant throughout the process [74]. If the stimulus changes shape, intensity, or presentation modality, the horse will react as in the presence of a new stimulus and the process of habituation will be invalidated. It appears that horses either adopt habituation (reduced behavioral response) or sensitization (increased behavioral response) to repeated exposure of a novel stimulus [22].

Through non-associative learning, the horse can also become habituated to pressure, provided that it is light (because more pressure results in greater discomfort, with a greater intensity of discomfort, it is less likely that the horse will become habituated) [59]. This introduces the risk of riding a horse on contact, which is pressure. (In order to communicate with the horse, we need to establish a low baseline of contact [light pressure]. The horse will accept a consistent, light pressure and consider this a new baseline.) However, it is essential to know the result that constant pressure on the mouth (or on other parts of its muzzle based on the bridle used) has on the horse. Without knowledge of non-associative learning and without the necessary attention to timing, a rider may not be able to adequately assess the extent of contact with the horse's mouth and without intent, can apply pressure of some intensity [75]. A rider could consistently apply a level of pressure similar to one the horse has associated with the stimulus to certain behaviors, but without consciously giving any command. As the work proceeds, the horse would then become habituated to this constant pressure on its mouth, creating a new, higher baseline of pressure, which would result in the pressure losing its effect [59]. The similar intensity of two different pressure stimuli (one of which is used for communication) is a risk because the horse may lose the ability to distinguish between them [34]. Thus, it may not be able to perceive the difference between the pressure signal associated with a specific behavior and the baseline contact with the mouth, which instead has no meaning [32].

THE REINFORCEMENT AND THE PUNISHMENT: A NOT WELL DEFINED BOUNDARY

As previously mentioned, negative reinforcement is the removal of an aversive stimulus (that creates discomfort) from the horse. Any aversive stimulus (including pressure implemented by humans) is nothing more than a punishment, even when light [29]. In the human mind, the word "punishment" conjures up negative scenarios, relating the use of whips or other means of coercion, but it should not be understood as simply inflicting physical pain or mental coercion. Light punishment is employed every time pressure is applied to the horse (*No, this is not what I want*), the removal of which (*Yes, this is what I want*) allows implementation of negative reinforcement. It is essential to highlight this concept because only awareness of our behavior allows us to apply negative reinforcement correctly. Through a total understanding of the difference between negative reinforcement and positive punishment, we see that if the pressure is not removed immediately after the horse performs the correct behavior, it would actually result in a punishment for that behavior by saying to the horse "*No, this is not what I want*". Therefore, the non-application of the reinforcement (failure to remove pressure) can be interpreted by the horse as a punishment [76] thus indicating how thin the line is between proper communication and sending contradictory signals. To eliminate an undesired behavior of a horse, it is important to know the psychological rules of the punishments and consequences to avoid errors in their application. The effectiveness of the punishment depends partially on its intensity. This does not mean that full strength should be used, but rather that the intensity should be suitable for that specific horse at that specific time (any horse, like any person, has a threshold at which the punishment is effective). The other critical element is the time interval between the behavior that needs to be punished and the administration of the punishment. The more time that passes between the two, the greater the chance that the punishment is ineffective and has only undesirable consequences. As for correct application of reinforcement, learning is effective only within a timeframe of a few seconds, outside of which it is likely that the punishment takes on a different meaning [29]. For example, if the horse is whipped following a refusal of a fence, it may not know whether to associate the stimulus with the refusal itself or with some other part of the ongoing situation. The horse could associate the punishment with the fence, the jumping, or even the riding. A very high percentage of punishments administered as such are incorrectly

applied regarding timing and intensity that is, without the consideration of all conditions in which punishments are unconsciously administered [76]. The punishment can quickly be associated with the person who performed it, and this will bring the horse to implement defensive or escape behaviors as soon as this person is present in the training process. In the same way, that association can be extended to places or objects [29]. Repeated punishment with the wrong timing of application [76] induces reduction of spontaneous activity, which is often associated with decreased attention and learning [77].

The reduction of spontaneous activity can lead to a negative perception of the environment even if the horse is stabled appropriately for its needs, consequently reducing the welfare state. These two conditions may result in pathological forms of apathy and an inability to adapt to the environment, that could lead horses toward depressive-like disorders [17, 18, 48]. Finally, the most dangerous effect induced by punishment is aggressiveness toward humans [29]. Therefore, for all the aforementioned reasons, positive punishment should be avoided as much as possible.

CONCLUSION

In conclusion, to ensure an adequate level of equine welfare, it is necessary that the psychological rules of animal learning are applied during training and management. This will lead to positive effects for all sports and activities in which the horse is employed. Furthermore, this will also reduce the possibility of injuries for humans involved in the equestrian industry, with a significant reduction of costs for civic society and the national health system. This review summarized the actual scientific and practical knowledge of learning theory to practically apply them in the field. These concepts could be easily taught; therefore, the authors suggest that all individuals who work or manage horses by profession or passion, should attend specific training programs on psychological rules of animal learning in order to apply this knowledge during their daily work with horses.

Acknowledgements

The authors are grateful to Cory Kieschnick (Equine Science Department, Delaware Valley College) and Ronald DePeter (Writing Center, Delaware Valley College, USA) for proofreading and English revision.

Conflict of interest statement

There are no potential conflicts of interest or any financial or personal relationships with other people or organizations that could inappropriately bias conduct and findings of this study.

Received on 12 May 2014.
Accepted on 24 February 2015.

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