THE POSTURAL SIGNATURE DEVICE: A NON-INVASIVE TOOL FOR IDENTIFYING MEDICATION THRESHOLDS. A PRELIMINARY REPORT

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ABSTRACT

A postural signature device (PSD) was developed for measuring behavioural responses in the horse after administration of therapeutic agents or medication. A force plate, placed under one hoof, monitors a horse's hoof balance. Vertical forces applied to different areas of the force plate are recorded every 50 milliseconds over a 1 min sampling period. Data are analysed statistically to demonstrate the horse's response to medication or therapeutic agents. The PSD is proposed as a tool for the early diagnosis of navicular syndrome which is reportedly characterised by uneven weight distribution on a hoof. Additionally, it is hoped that the technique will be of use in determining the therapeutic window or 'no-effect' threshold of medications which affect the locomotion or performance of horses.

INTRODUCTION

The use of therapeutic medication by veterinarians is often necessary to protect the health and welfare of horses in training. However, these same medications have the potential to alter the performance of racehorses. In order to ensure fairness in competition, post race blood or urine testing is performed routinely on racehorses. However, the currently available sensitive methods of detection may identify traces of such agents administered days before the event, or amounts that are well below therapeutic or 'no-effect' concentrations. (Tobin *et al.* 1999). Various physiological responses have been used for determining the highest no effect doses or concentrations of these agents. These include step counting, hoof withdrawal reflex latency, skin twitch reflex latency, penile protrusion, variable interval conditioning, physiological variables, head ptosis (Harkins *et al.* 1994) and behavioural chamber activity (Harkins *et al.* 1996a). Comparison of results obtained from a variety of testing methods is useful, as different techniques may be applicable to specific compounds or agents (Harkins *et al.* 1996b, 1997).

As early as 1873 Marey (1873) equipped horses with accelerometers to record gait parameters. Since then, numerous devices have been used to record either dynamic or static locomotor activity. The kinetic devices are either affixed to the horse in the manner of horse shoes (Frederick and Henderson 1970; Barrey 1990; Ratzlaff *et al.* 1990) or are placed on the floor with the horse standing on them (Aviad 1988; Barrey 1999). They can be used both to record data that are not readily clinically observable, and to obtain objective data without relying on the subjectivity of human observations. In this regard, we have chosen to develop and test a postural signature device (PSD), which includes a standing force plate, because of its potential to obtain measurements from different horses without the need to alter their shoes.

MATERIALS AND METHODS

Seven mature Thoroughbred mares (6 sound and one lame) weighing about 500 kg, were used. They were fed twice daily on grass hay and feed (12% protein), as a 50:50 mixture of oats and alfalfa-

Fig 1: Top view of the loaded force plate of the PSD.

based protein pellet. They were vaccinated annually for tetanus (Tetanus Toxoid, Fort Dodge Inc., Iowa, USA) and de-wormed quarterly with ivermectin (MSD Agvet, New Jersey, USA). A routine clinical examination was performed before each experiment. The horses were managed according to the rules and regulations of the University of Kentucky's Institutional Animal Care Use Committee, which also approved the experimental protocol.

The postural signature device

The postural signature device consists of 2 (200 x 200 x 25 mm) steel plates on which the horse stands (the force plate; Fig 1), electronic devices that measure the pressure signal and a computer that records the generated weight data. Weight is measured by 3 load cells sandwiched between the 2 steel plates. This device was developed with the aid of computer assisted design software (Autocad 2000, Autodesk Inc, California, USA). The thickness of the steel plate ensures a pressure induced deformation of less than 10% which is required for accurate data transmission to the load cells.

The upper surface of the plate was marked with a 40 mm grid, which was used for positioning the hoof on the force plate. The dorsal part of the toe was placed on the first latero-medial line and the quarters symmetrically positioned along the lateral and medial lines of the plate. Four adjustable feet were used to level the plate if necessary. If the horse moved during testing the grid allowed the hoof to be re-positioned easily.

The load cells (type L2761, Futek, California, USA), in the shape of 'washers', had a capacity of 226.8 kg each. They were 12.7 mm thick with internal and external diameters of 9.53 and 37.60 mm, respectively. Three vertical steel rods welded to the top plate slid freely through the washers and the bottom plate, thereby providing support and positioning for the load cells. Two rods were located at the palmar corners of the plate while a third rod was placed at the mid-point of the dorsal side of the plate. The load cells were powered with 10 V DC through 4 amplifiers (type JM-2, Futek, California, USA). A transformer (type HA15-1.5-A, Power-one International, California, USA) provided 15.2 V DC for the remaining devices.

The amplifier modules served 3 functions: load cell calibration, power supply and signal amplification. The amplified signal was measured with a voltmeter (CB-7017, Computerboards, Massachusetts, USA). The load cells produced a continuous signal from which the voltmeter sampled 20 readings/s. Signals were transferred through a serial communication device (CB-7520, Computerboards, Massachusetts, USA) to a laptop computer where they were recorded using a Visual Basic program (Visual Basic 5.x, Microsoft, Washington, USA) running under Windows 95. The program transmitted a data file every 0.05 s. Using a macro, these data were transferred directly into a spreadsheet (Excel 2000, Microsoft, Washington, USA) with each row corresponding to a time measurement set.

Calibration of the load cells and voltmeter

The load cells were not calibrated individually, but as an integrated unit in the force plate. A variable load was applied (Fasttrack, Instron, Massachusetts, USA) at the centre of the triangle formed by the 3 load cells. For each cell, a regression line was computed using the general-purpose procedure for regression of SAS (PROC REG, SAS 8.00, SAS Institute Inc, North Carolina, USA) to give the relationship between the load applied and the output voltage. The voltmeter was calibrated according to Computerboards' recommendations by iteratively applying 0 and 5 V DC.

Data recording and processing

Data files were composed of either one or many readings interspersed with aberrant values due to non-loading or mis-loading of the plate. Values related to rapid non-loading and re-loading appeared as values exceeding the 0-5 V range with very large standard deviations. Values related to mis-loading are characterised by absent or extreme low values from one cell and values nearing the

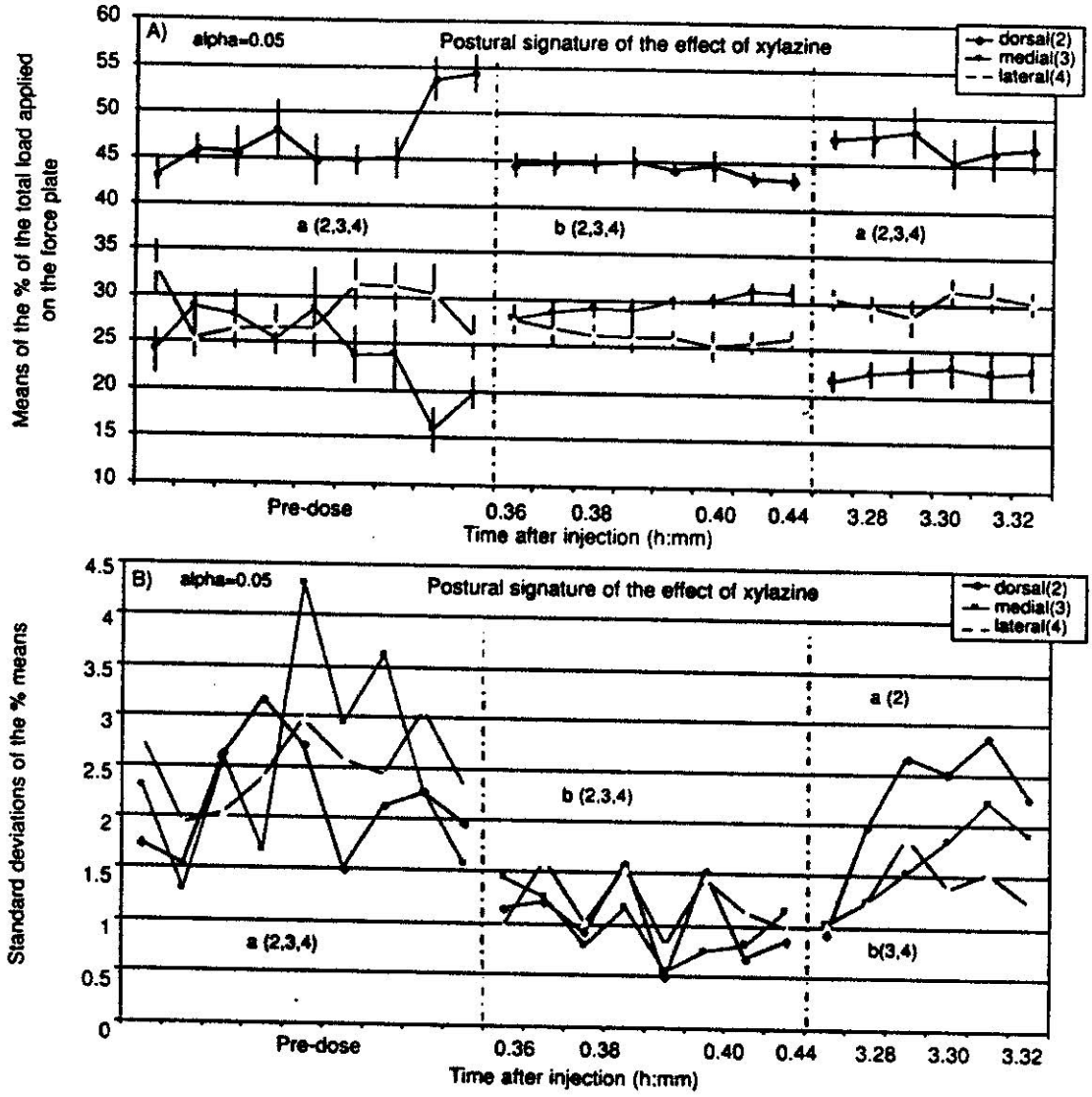

Fig 2: Postural signature of the effect of xylazine: A, % of mean of total load per hoof measured before and after injection - B, standard deviation of measurements in part A - 2, 3 and 4 refer to dorsal, medial and lateral curves, respectively - a, a notation indicates 2 sets of data having no significant difference - a, b notation indicates 2 sets of significantly different data therefore, a(2), b(2) indicates 2 significantly different sets of dorsal (2) data

maximum from another cell. The Excel spreadsheet data file was opened and all aberrant values removed, leaving sets of data composed of about 1.200 measurements per minute.

Raw data were recorded in the form, 'time(yyyy-mm-dd hh:mm:ss), channel-2 voltage, channel-3 voltage, channel-4 voltage (range of 0.000 to 5.000V)'. Using Excel's 'mid' function, time in the form 'hh:mm:ss' was extracted from the raw data. The difference between each time point and the time of drug injection was computed.

For each time point each load cell's regression line equation of voltage vs. load was used to calculate the 'mean load', the 3 loads summed to obtain 'total load' and the contribution of individual loads to the total load calculated as a percentage. For each set of data, the means and standard deviations of those percentages were computed.

Administration of medications

The postural signature device was used to collect data after the administration of 3 different drug treatments - 2 depressant treatments and one local anaesthetic.

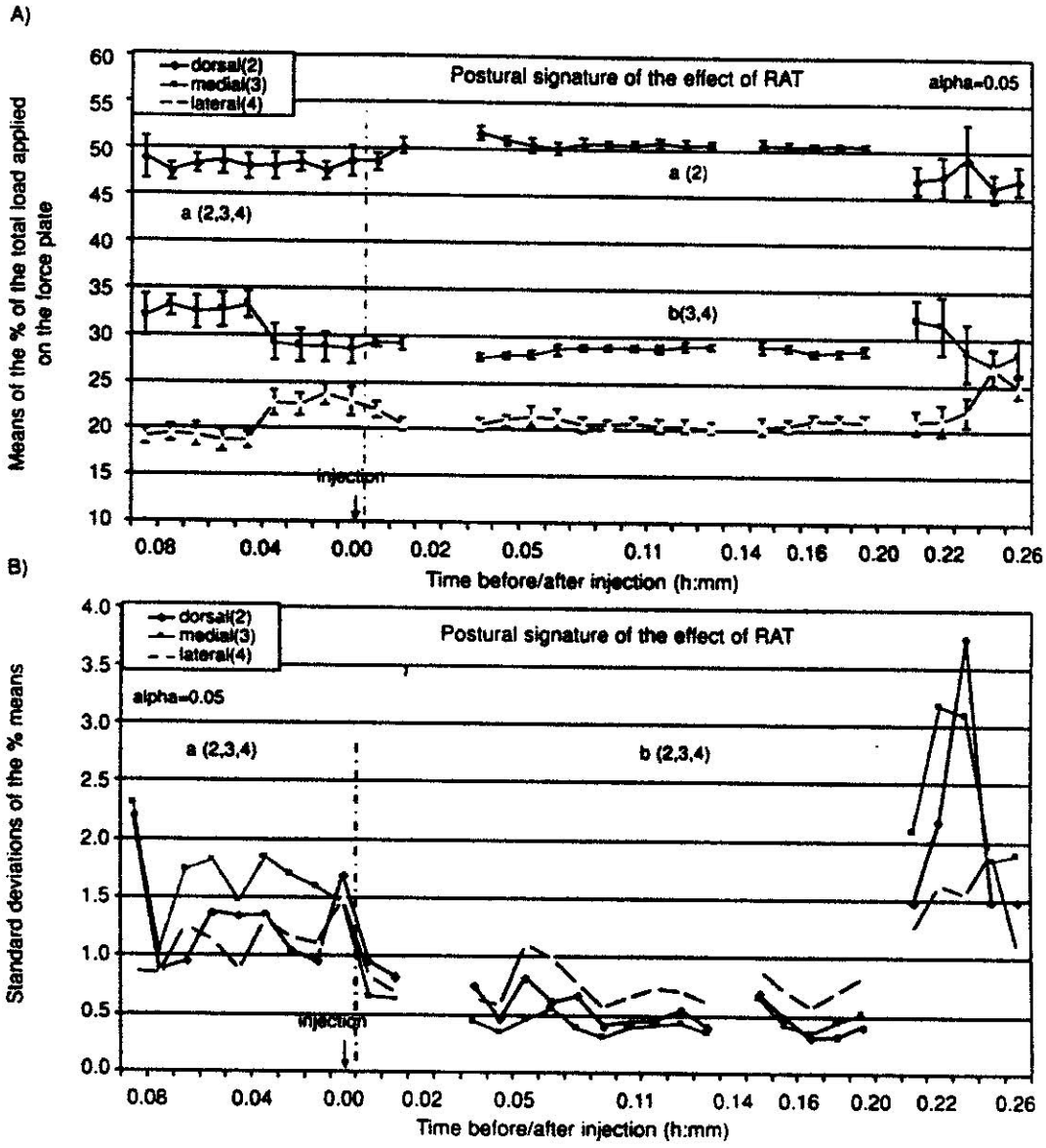

Fig 3: Postural signature of the effect of RAT : - A, % of mean of total load per hoof measured before and after injection - B, standard deviation of measurements in part A - 2, 3 and 4 refer to dorsal, medial and lateral curves, respectively - a, a notation indicates 2 sets of data having no significant difference - a, b notation indicates 2 sets of significantly different data therefore, a(2), b(2) indicates 2 significantly different sets of dorsal (2) data.

1) Xylazine hydrochloride (Rompun, Bayer, Leverkusen, Germany) was administered iv at 0.8 mg/kg, to 3 sound horses.

2) A mixture of xylazine hydrochloride (Rompun, Bayer, Leverkusen, Germany) at 0.8 mg/kg, acepromazine maleate (PromAce, Fort Dodge Inc, Iowa, USA) at 0.4 µg/kg and butorphanol tartrate (Torbugesic, Fort Dodge Inc, Iowa, USA) at 0.4 µg/kg was administered iv to 3 sound horses. This mixture is called RAT, an acronym for Rompun, Acepromazine and Torbugesic.

3) A horse diagnosed with chronic degenerative joint disease (DJD) of the knee was given 2 intra-articular injections (0.4 mg/kg) (palmar and dorsal approach) of mepivicaine hydrochloride (Carbocaine-V, Pharmacia & Upjohn, Michigan, USA).

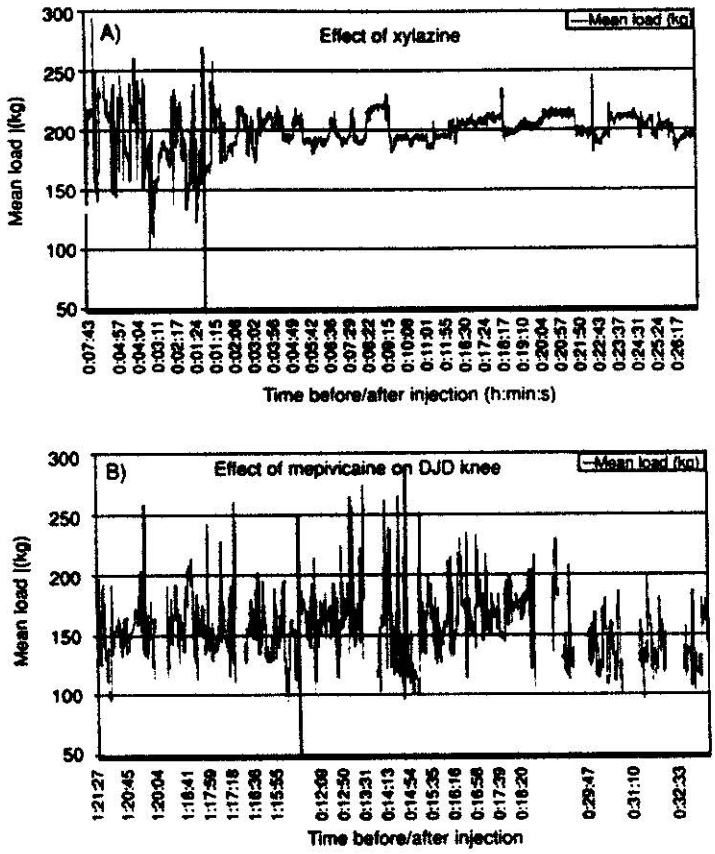

Fig 4: Comparison of depressant (A) and local anaesthetic (B) on mean load as measured by the postural signature device - A. xylazine effects, before and after treatment of a normal horse - B, mepivacaine effects, before and after treatment by intra-articular block of a horse with DJD.

Data analysis

Data were analysed statistically by the repeated measurements variance procedure of SAS. The baseline data obtained before injection of the drug were compared to the post injection data, and also to data obtained in the late post injection (post effect period), when the drug was considered to be pharmacologically ineffective. All comparisons were made at a = 0.05.

RESULTS

Effect of xylazine

Minutes after the iv injection of xylazine, the horse became quieter, moved less, took profound inspirations and gradually lowered its head. Figure 2 shows the means (A) and standard deviations (B) of the means of the percentage load applied on each load cell before and after administration of xylazine.

Figure 2A shows significant differences in means between the period under influence of xylazine and both pre-dose and post effect period (about 3.5 h later). Data from the pre-dose and post effect periods are not significantly different. The data obtained from each channel are in good agreement.

However this is not the case in Figure 2B. The palmar data (channels 3 and 4) show a significant difference in standard deviations between the pre-injection and post injection periods, but the post effect period data do not differ significantly from that obtained during the period of maximum action (35 to 45 min after the injection) of the drug (Fig 2B). However, the dorsal standard deviations data (channel 2) agree with respect to the means and standard deviations; ie the pre-dose and post effect period are not significantly different, but are both significantly different from the post dose period.

Effect of the depressant cocktail RAT

The iv injection of RAT induces the same clinical signs as xylazine itself. Figure 3 shows the means and standard deviations of the means of the percentage load applied on each load cell before and after administration of the depressant cocktail RAT. No post effect period data were recorded. There is no significant pre- and post dose difference in the percentage mean of the load

applied to the dorsal load cell. Except for the dorsal means data, all the values (means and standard deviations) of post injection data are significantly different from the pre-injection data.

Effect of the intra-articular block

Although the clinical signs of degenerative joint disease (DJD) decreased following the intra-articular injections, analysis of the postural signature data failed to demonstrate any hoof balance response to the intra-articular block. The computed data: total load, mean load, and percentages did not reflect a consistent change in the behaviour of the horse, although total hoof weight placement on the hoof was slightly different (data not presented).

Depressants (Fig 4A) induced a PSD mean load response clearly different from that of local anaesthetics (Fig 4B). The effect of local anaesthesia on a knee affected with degenerative joint disease is apparently more readily detected by visual observation of the horse trotting than by the PSD foot balance data.

Discussion

Based on the data presented here, some interpretations may be advanced concerning the value and potential uses of the postural signature device. Data interpretation also suggests there are some technical problems to be solved. As shown in Figure 4A, the effect of depressants on the measured variables was clear-cut and readily observable. However, the ANOVA data analysis on percentage means was not particularly effective for distinguishing between pre- and post drug administration values, because of the very large baseline variability recorded. This baseline variability is due to spontaneous activity of the horses, and this makes identification of drug effects more difficult. Potential solutions to this problem include the use of multicomponent analysis or neural network analysis, and preliminary work in this area is very promising.

The demonstrated value of the PSD in distinguishing the effects of depressants on locomotor passive reflexes suggests that this device might be used for estimating threshold limits for the effects of certain depressant drugs. This tool may be more useful than the head ptosis method as it relates more closely to locomotor system activity. Urine and plasma sampled simultaneously with acquisition of postural signature data could be used for setting up threshold limits by measuring the concentration of drugs and metabolites in plasma and/or urine. Pharmacokinetic endpoints could be compared to the time points at which the post dose data no longer differ from the pre-dose data.

A problem in setting individual drug thresholds arises where synergistic effects may occur from the use of a drug mixture. This problem is not inherent to the PSD but instead arises from the method used by jurisdictions for setting thresholds. The great variety of threshold limit values in different racing jurisdictions points to the inherent problems in setting up uniform thresholds. The addition of new data, with a method different from those previously used, could lead to consistency.

While the PSD has only been used to investigate the action of depressants, it could also be used for other groups of drugs. For stimulants, the reduction of the recording time to a fourth of the length used for depressants should still give valid and useful results.

Although the PSD data failed to demonstrate a significant behavioural response to a local anaesthetic in the DJD affected horse, it may nevertheless be useful for detecting lameness associated with a foot. For example, navicular disease has an aetiology that, although still uncertain (Gabriel *et al.* 1997), might arise from biomechanical considerations (Wright 1993). Assessment of the balance weight of the foot may be useful in treatment of navicular disease (Ostblom *et al.* 1984; Stashak 1996). The best value of the PSD might be in measuring the balance of the foot before clinically obvious lameness occurs, as a predictive tool, or after treatment to evaluate drug or treatment efficacy, for example, validating the effects of remedial shoeing.

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