

## Quantification of functional behavior in humans and animals: Time for a paradigm shift

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**Abstract**—Measuring the effectiveness of interventions aimed at restoring motor function will be critical in deciding which animal trials should be translated to human studies. While various methods of quantifying motor behavior exist, many of these rely on observation and interpretation. Kinematic analysis is an objective means of quantifying temporal relationships and coordination within and between limbs during motor performance. These relationships offer valuable information on the condition and organization of the underlying neural circuitry. Kinematic data can drive a number of powerful analyses. These analyses can help detect nuances in motor performance and answer questions about the effectiveness of interventions designed to restore motor function.

**Key words:** coordination, interlimb, intralimb, kinematics, motor control.

### INTRODUCTION

Damage to the spinal cord results in disrupted physiology at all levels of function, from the cellular to the systemic. Of all the detrimental effects of spinal cord injury (SCI), one of the most devastating is the disruption of the ability to perform functional movement. To evaluate the effectiveness of any intervention, one must measure a change in function; for this reason, the emphasis must be on quantifying functional motor behaviors. The

measurement of movement requires that observations be assigned numeric values.

Over the years, a number of useful tools have been developed to measure movement. However, the process by which numbers are assigned to the observation dictates how the outcome may be interpreted and what types of statistical tests can be applied to the data. The simplest scale of measurement is the nominal scale; movement classified with this type of scale typically poses a question that has a dichotomous response, such as whether a particular behavior (for example, the placing response) is present or absent.

The ordinal scale is superior to the nominal scale and may involve observational analysis, such as when a rater

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**Abbreviations:** BBB = Basso, Beattie, Bresnahan scale, SCI = spinal cord injury, SCI-FAI = Spinal Cord Injury Functional Ambulation Inventory, WISCI = Walking Index for Spinal Cord Injury.

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evaluates qualitative aspects of an animal's movement following SCI. The observations are assigned values that have a relative order, but the interval between values may not be equal. Examples of such scales developed for use in animals are the Basso, Beattie, Bresnahan Scale (BBB) [1] and the modified Tarlov scales [2]; and, in humans, the Spinal Cord Injury Functional Ambulation Inventory (SCI-FAI) [3] and the Walking Index for Spinal Cord Injury (WISCI) [4]. In most instances, it is not appropriate to apply parametric statistics to nominal or ordinal data.

Interval and ratio scales are the highest scales of measurement. These scales are amenable to analysis with parametric statistics. Interval scale measures have equal distances between observations and thus may be added or subtracted. However, because this scale lacks an absolute zero, the values may not be multiplied or divided. Temperature ( $^{\circ}\text{C}$  or  $^{\circ}\text{F}$ ) is the most common example of interval scale measurement. Ratio scale measures do possess a true zero, and therefore are amenable to all forms of mathematical operations. Many motor performance-related measures, such as speed, time to execute a task, number of errors, footprint analysis, phase relationships, swing-to-stance ratios, and force production, fall into the category of ratio scale data.

Given this broad array of powerful measures, it seems logical to select either an interval or ratio scale for quantification of movement. But which of these gives us the best information about the ability of the nervous system to control movement?

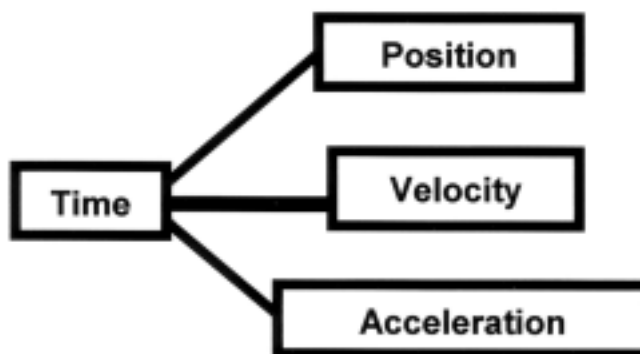
The premise of this article is that the most precise means of assessing neural control over motor output is kinematic analysis of the movement that is produced by the human or animal. In the context of this review, kinematic analysis is the quantitative description of the movement of the limbs, without regard for the forces that produced them. In limbed animals, the temporal relationships within a limb and between limbs reveal important information about the organization of the nervous system and its ability to control the limbs and thereby produce functional movement. Measures of motor output, such as muscle strength and speed, are important only to the extent that they contribute to function (unless one is a weightlifter or sprinter). The analysis of limb movement reveals information about the integrated function of all the physiologic systems involved in motor control. The principles of kinematic analysis are broadly applicable across all species of limbed animals (and to some extent, to nonlimbed animals as well), despite anthropometric differences.

Movement is characterized by such linear and angular kinematic variables as displacement and its time derivatives, velocity and acceleration. Kinematic analysis is an expansive topic; this article focuses only on a few selected techniques that are well suited to the types of behaviors that are studied in both humans and animal models.

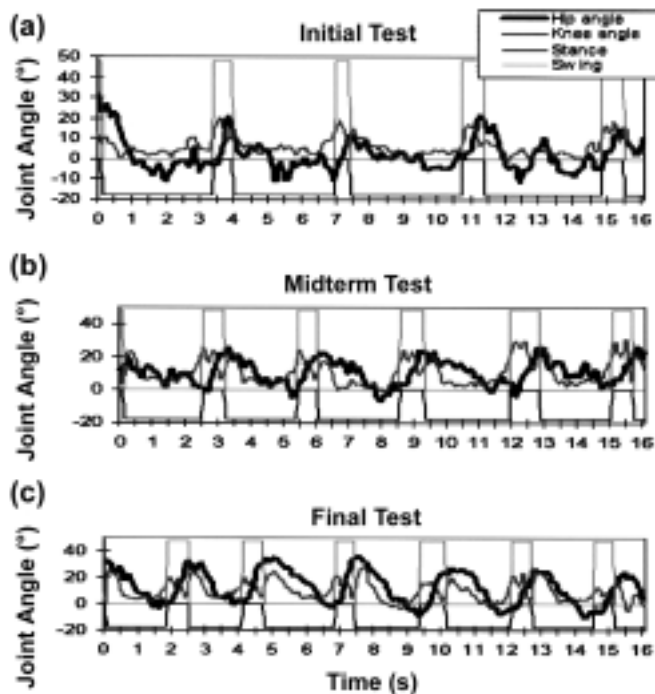
## TIME-SERIES OR TEMPORAL MOTION ANALYSIS

Among the earliest studies of animal and human movement are those of Eadweard Muybridge ... [5] and of Etienne-Jules Marey [6], who made extensive studies of walking on the basis of sequential still photographs in the mid- to late 1800s. Among the earliest and most systematic modern studies of animal movement are those of Erich von Holst [7], who made extensive studies of animal movement. It was the strict periodicity of these movements that led him to conceive the theory of the neural oscillator.

Plotting motion (displacement, speed, or acceleration) against time (**Figure 1**) is one of the most common techniques for evaluating limb movement. Time-series plots (**Figure 2**) are conducive to conveying the range and regularity of motion over the capture period and have been used for decades. Forssberg et al [8,9] were among the first to use these plots to illustrate interlimb and intralimb coordination in the spinal cat. These data are useful for identifying specific parameters, such as the mean joint angle at a specific point in the locomotor cycle, or for determining the range of joint movement. Such plots, the foundation of kinematic analysis, are suitable for evaluating change in a behavior, but they are not



**Figure 1.** Potential relationships in time-series analysis.



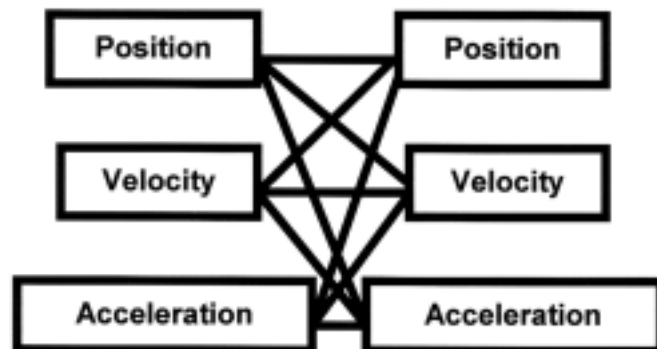
**Figure 2.** Time-series plots of hip and knee displacement at (a) beginning, (b) mid point, and (c) end of locomotor training program.

well suited for quantifying relationships among or between behaviors.

## RELATIVE MOTION ANALYSIS

Relative motion plots illustrate the displacement, velocity, or acceleration of one limb segment relative to another limb segment; therefore, time is removed from the representation (**Figure 3**). These representations are also called cyclograms or angle-angle plots when the plotted parameters are displacement versus displacement. Phase-plane (or state-space) plots are an alternative form of relative motion plot, in which the velocity or acceleration of a limb is plotted against its displacement. Riley et al [10] hypothesized that postural stability requires control of both position and momentum of the center of gravity, and they used velocity versus displacement plots to characterize balance control in subjects with vestibular hypofunction and in unimpaired individuals.

Analysis of relative motion is particularly well suited to assessing the stability of a behavior over multiple trials and therefore to evaluating coordination. In the Motor

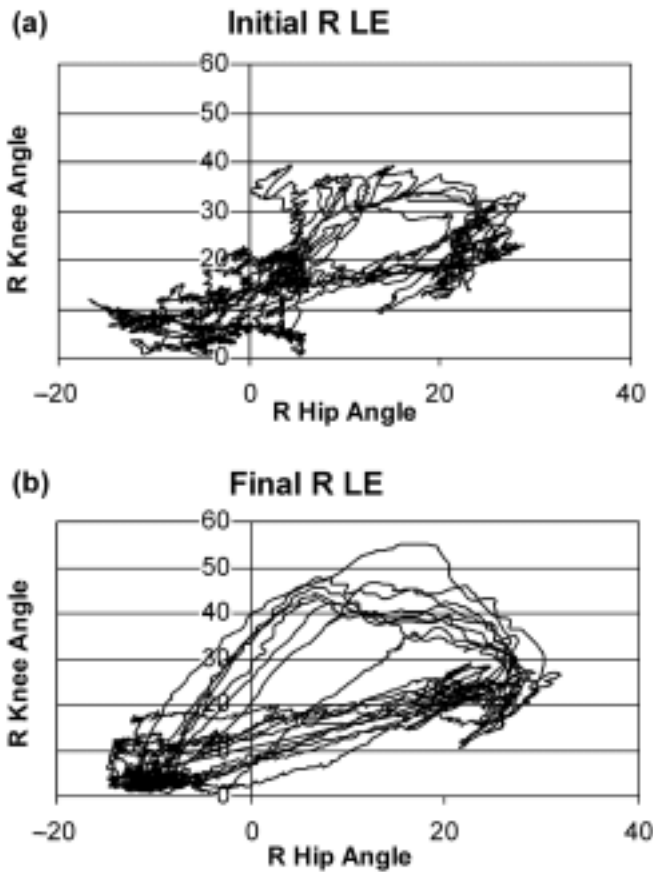


**Figure 3.** Potential relationships in relative motion analysis.

Rehabilitation Laboratory at The Miami Project to Cure Paralysis, we use these plots to assess limb coordination in individuals with SCI who participate in locomotor training studies (**Figure 4**). We recently developed a vector coding technique [11] to quantify relative motion plots and thereby compare the degree of agreement (or correspondence) between multiple gait cycles, pre- and post-training [12]. This technique is a refinement of Freeman's encoded chain technique [13], which was developed with the intent of describing the outline of figures on a video monitor in terms their relative pixel position.

The vector coding technique treats each frame-to-frame interval as a vector (i.e., having both direction and magnitude), defined by the change in the  $x$ -direction (e.g., hip displacement) and the change in the  $y$ -direction (e.g., knee displacement). If the vector joining frames 1 and 2 of the first cycle of a repetitive behavior (e.g., step cycles) has the very same direction as the vector joining frames 1 and 2 of the second cycle, and if this is true of all frame-to-frame intervals in the two cycles, then the relative motion plots for cycles 1 and 2 will have the same shape. The vector defined by the hip-knee trajectory between frames 1 and 2 in cycle 1 can be compared to the same interval for cycle 2, cycle 3 ... cycle  $n$ . To quantify this relationship, the difference between frames 1 and 2 for the hip angle values ( $x_{1,2}$ ) and the knee angle values ( $y_{1,2}$ ) is calculated. These values represent the change in the  $x$ - and  $y$ -directions, respectively, in the frame-to-frame interval, between frames 1 and 2. The angular direction of the line segment,  $l_{1,2}$ , between two consecutive points or frames is calculated with the formula

$$l_{1,2} = \sqrt{(x_{1,2})^2 + (y_{1,2})^2} \quad . \quad (1)$$



**Figure 4.** Angle-angle plots for knee-hip coupling during overground walking in subject with SCI (a) before and (b) after participation in locomotor training program.

The cosine and sine of  $l_{1,2}$  are then found with the formulas

$$\cos \theta_{1,2} = x_{1,2}/l_{1,2} \quad , \quad (2)$$

and

$$\sin \theta_{1,2} = y_{1,2}/l_{1,2} \quad . \quad (3)$$

This process is repeated for each frame-to-frame interval within each cycle. The mean cosine ( $\cos \bar{\theta}$ ) and sine ( $\sin \bar{\theta}$ ) for a given frame-to-frame interval over multiple cycles (e.g., frames 1–2 of cycles 1– $n$ ) are calculated, and the mean vector length for that frame-to-frame interval is then determined with the formula

$$\sqrt{(\cos \bar{\theta}_{1,2})^2 + (\sin \bar{\theta}_{1,2})^2} = a_{1,2} \quad . \quad (4)$$

The length of the mean vector,  $a$ , denotes the degree of dispersion (or, conversely, of concentration) of the knee/hip values about the mean over multiple cycles for that particular frame-to-frame interval. The larger the value of  $a$  (between 0 and 1), the less variable (i.e., less randomly distributed, more consistent) is the hip/knee relationship.

The arithmetic average,  $\bar{a}$ , of all the mean vector lengths is found by

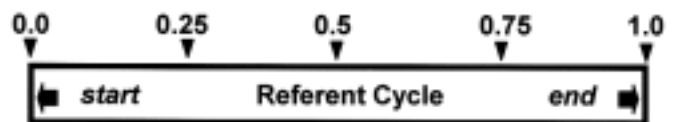
$$(a_{1,2} + a_{2,3} + a_{3,4} \dots a_{N-1,N})/N = \bar{a} \quad , \quad (5)$$

where  $N$  is the number of frames per cycle, and  $\bar{a}$  is the angular component of the coefficient of correspondence, which signifies the overall variability of the knee/hip relationship for all included cycles. If the relative motion between the hip and the knee is in perfect agreement over multiple cycles, then  $\bar{a} = 1$ , indicating maximal consistency between cycles for the shape of the plot (i.e., trajectories of the knee/hip relationship), but not necessarily the area (i.e., step lengths). Additional details on this analysis, as well as information for evaluating agreement between cycles relative to the area of the plot, are given elsewhere [11].

## PHASE ANALYSIS

In addition to plotting the angular displacement, velocity, or acceleration, features of movement can be characterized and quantified in terms of the proportion of the cycle (i.e., the phase) at which they occur. Phase analysis has an advantage over time-series and relative motion plots, as the process results in normalization of the data. This facilitates comparisons both within and between subjects.

The most usual type of phase analysis is single-referent phase analysis (**Figure 5**). This type of analysis requires the selection of a starting and ending point for a single “referent” cycle period, to which another event of interest is referenced. The starting point of the cycle is



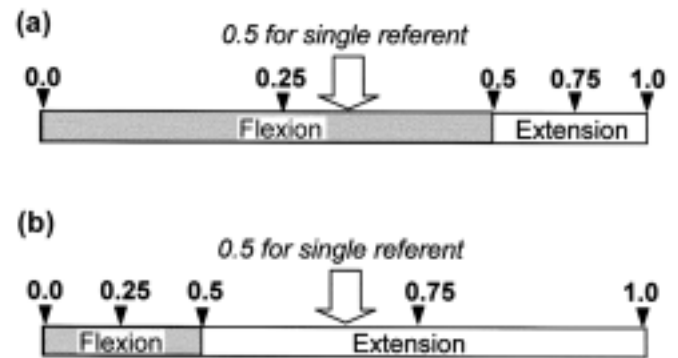
**Figure 5.** Phase values in single-referent phase analysis.

assigned a value of 0, and the end point a value of 1 (or  $0^\circ$  to  $360^\circ$ , in the case of a polar plot). In the analysis of walking, the onset of heelstrike is most commonly used to designate the start/end of the period. But this is an arbitrary selection, and any obvious event in the cycle (e.g., onset of hip flexion) could be used just as well. The latency of an event of interest (e.g., contralateral heelstrike) is the time between the start of the referent cycle and that event of interest. The phase of the event of interest is defined as the proportion of the reference cycle period at which the event of interest occurs, or

$$\text{Phase value of event of interest} = \frac{\text{latency of event from onset of cycle period}}{\text{cycle period}}. \quad (6)$$

An event of interest that occurs in the middle of the referent period, therefore, has a phase value of 0.5; while an event that occurs at the start (or end) of the cycle has a value of 0 (or 1). A recent paper by Kullander et al [14] used circular plots to demonstrate abnormal locomotor coordination in transgenic mice. The alternating hindlimb behavior (i.e., relative interlimb phase = 0.5) usually observed for walking in wild-type mice was replaced by synchronous behavior (i.e., hopping; relative interlimb phase = 1) in mice in which a receptor or ligand important for function of the locomotor central pattern generator had been genetically altered.

Single-referent phase analysis is very useful for comparing similar forms of behavior. However, there are instances in which one is interested in documenting similarities between different types of behaviors. Dual-referent phase analysis is very useful for comparing different forms of behavior that have similar components (such as a flexion component and an extension component), but in which different proportions of the cycle period are occupied by each component. For example, in the rostral scratching behavior of the spinal turtle, the hip extension component has a longer duration than the hip flexion component; in the pocket scratching behavior, however, the hip extension component has a shorter duration than the hip flexion component [15]. Dual-referent phase analysis allow the investigator to determine the timing of an event of interest (such as the onset of knee extension) relative to the period of the component during which the event occurs (**Figure 6**). With the use of a mathematical adjustment, the phase of the event of interest is calculated relative to the period of the component, rather than to the entire cycle period, as follows:



**Figure 6.** Phase values in dual-referent phase analysis (with single-referent analysis value indicated by open arrow): (a) cycle with long flexion component and (b) cycle with long extension component.

$$\text{Phase value of event occurring during flexion component (FC)} = \frac{\text{latency of event from FC onset}}{(2 \times \text{FC period})}. \quad (7)$$

$$\text{Phase of event occurring during extension component (EC)} = 0.5 + \left[ \frac{\text{latency of event from EC onset}}{(2 \times \text{EC period})} \right]. \quad (8)$$

As such, an event that occurs at the time of the transition between the flexion and extension always has a phase value of 0.5, an event that occurs at the midpoint of the flexion always has a phase value of 0.25, and an event that occurs at the midpoint of extension always has a phase value of 0.75. Field and Stein [15] used this type of analysis to discern similarities in knee extension onset times between scratching behaviors in spinal turtles and swimming behaviors in intact turtles. Dual-referent phase analysis allows for meaningful comparisons of the similarities between these behaviors that would not be apparent with single-referent phase analysis.

## SUMMARY

Kinematic data may be analyzed in a variety of ways, and the selection of a particular analysis is based largely on the research question being investigated. Time-series plots are useful to assess the range of movement and obtain an impression of the behavioral pattern exhibited over time. Relative motion analyses remove time as a variable and allow direct appraisal of the consistency of the behavior being studied. The degree of consistency can be quantified with vector coding techniques. Finally,

phase analyses allow for comparisons of a behavior between animals or between individuals. This form of analysis is also useful for comparing different behaviors and different forms of a behavior. The selection of single-referent versus dual-referent phase analysis depends on whether the investigator is interested in the timing of a particular event relative to the cycle as a whole, or relative to the components of the cycle. Whatever type of kinematic analysis is used, the quantification of motor behaviors is the most incisive way to assess motor function. As such, these forms of analysis provide a definitive means to critically evaluate the effectiveness of interventions aimed at restoring motor function following SCI.

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