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RESEARCH ARTICLE

# Forelimb kinematics during hopping and landing in toads

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## ABSTRACT

Coordinated landing in a variety of animals involves the re-positioning of limbs prior to impact to safely decelerate the body. However, limb kinematics strategies for landing vary considerably among species. For example, human legs are increasingly flexed before impact as drop height increases, while turkeys increasingly extend their legs before impact with increasing drop height. In anurans, landing typically involves the use of the forelimbs to decelerate the body after impact. Few detailed, quantitative descriptions of anuran forelimb kinematics during jumping exist and it is not known whether they prepare for larger landing forces by changing forelimb kinematics. In this study, we used high-speed video of 51 hops from five cane toads (*Bufo marinus*) to test the hypothesis that forelimb kinematics change predictably with distance. We measured excursions of the elbow (flexion/extension) and humerus (protraction/retraction and elevation/depression) throughout every hop. The results indicate that elbow and humeral excursions leading up to impact increase significantly with hop length, but do so without any change in the rate of movement. Instead, because the animal is in the air longer during longer hops, near-constant velocity movements lead to the larger excursions. These larger excursions in elbow extension result in animals hitting the ground with more extended forelimbs in longer hops, which in turn allows animals to decelerate over a greater distance.


**KEY WORDS: Anuran, Jumping, Deceleration, Control**

## INTRODUCTION

Landing is an inevitable consequence of jumping. Coordinated landing requires energy dissipation over relatively long time periods, and in many vertebrate jumpers, limbs are used to extend the distance and time over which animals decelerate and dissipate energy after impact. A survey of kinematic strategies for landing suggests that limbs may be used in quite different ways across taxa to prepare for the moment of impact. For example, humans increasingly flex their legs as drop height increases, leading to a more flexed limb at impact during high drops (Ford et al., 2011; Hsu and Huang, 2002; Peng et al., 2011; Santello et al., 2001). In contrast, turkeys (Konow and Roberts, 2015) increasingly extend their legs with drop height, leading to more extended limbs at impact during larger drops. Additional studies of cats (McKinley and Smith, 1983) and monkeys (Dyhre-Poulsen and Laursen, 1984), did not emphasize effects of jump or drop distance on impact preparation kinematics, although both studies suggest that landing limbs are nearly fully extended at the point of impact.

In anurans, the clade of vertebrates perhaps best known for using jumping as a primary means of locomotion, descriptions of limb kinematics in preparation for impact have remained largely qualitative. Several studies have shown that various anuran species prepare for high landing forces by moving their forelimbs anteriorly during the aerial phase of a jump to help brace for impact (Gillis et al., 2010; Griep et al., 2013; Nauwelaerts and Aerts, 2006; Peters et al., 1996). The degree to which the forelimbs are then able to decelerate the body varies markedly among species.

Cane toads, *Bufo marinus* Linnaeus 1758, have recently received considerable attention as a model for studying the control of landing (Gillis et al., 2010; Akella and Gillis, 2011; Azizi and Abbott, 2013; Ekstrom and Gillis, 2015) because unlike more basal anurans, which often crash-land head or trunk first (Essner et al., 2010), cane toads routinely perform controlled, coordinated landings. To enable coordinated landing, they position their body and limbs appropriately in mid-air so as to align the ground reaction force vector close to the center of mass (Azizi et al., 2014), allowing them to balance on their forelimbs momentarily as they lower their hindlimbs to the ground (Akella and Gillis, 2011; Gillis et al., 2010; Griep et al., 2013). Such control prepares animals well for the next hop, and may enable their migratory capability by allowing them to effectively string together many short hops to cover large distances (Estoup et al., 2010; Phillips et al., 2007). A recent review focusing on landing behavior in cane toads suggests that landing preparation may be complex (Gillis et al., 2014). Like in humans, monkeys and cats (Magalhães and Goroso, 2009; McKinley and Smith, 1983; Santello and McDonagh, 1998), cane toads increase the intensity of pre-landing recruitment in antagonistic forelimb muscles in longer hops (Gillis et al., 2010; Ekstrom and Gillis, 2015), in preparation for greater impact forces (Nauwelaerts and Aerts, 2006).

Given this distance-dependent pattern of pre-landing forelimb muscle recruitment in toads, associated forelimb movements might also change with hop length. Yet, previous examinations of anuran forelimb kinematics have not addressed this question, instead emphasizing important features of the landing event itself (Griep et al., 2013; Nauwelaerts and Aerts, 2006), the role of pectoral girdle anatomy (Emerson, 1983; Griep et al., 2013) or more general kinematics of the hop cycle (Peters et al., 1996). Recent work by Azizi and Abbott (2013) suggests that elbow excursions change with hop distance in *B. marinus*. During toad hopping, shortening and lengthening strains in the m. anconeus, a monoarticular elbow extensor, increase with hop length as the elbows extend before impact and flex after impact, respectively. Azizi and Abbott (2013) argue that these changes in fascicle strain before and after impact likely parallel one another and are important for preventing injuries associated with overstretching muscles involved in dissipating energy during landing (Azizi and Abbott, 2013). While one can infer distance-dependent elbow excursions based on these patterns of fascicle length change, such excursions have not been measured directly. A full kinematic description and analysis of forelimb movements during jumping in toads will improve our understanding of whether they alter limb kinematics in a way that helps to prevent muscular damage during landing.

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**Table 1. Mean values of α, β and δ at the start and end of each phase**

| | $T_0$ | $T_1$ | $T_2$ | $T_3$ | $T_4$ |
|---|---|---|---|---|---|
| Elbow extension, α (deg) | 79±14 | 90±12 | 61±6 | 108±14 | 69±9 |
| Humeral protraction, β (deg) | 49±17 | 37±12 | 64±10 | 109±12 | 54±8 |
| Humeral depression, δ (deg) | 172±11 | 175±11 | 174±8 | 137±11 | 166±12 |

$T_0$–$T_4$, time points defining the beginning and end of each phase.

If toads modulate forelimb kinematics with distance, this analysis could also reveal how such modulation is achieved, giving us further insights into the control strategies involved. If forelimbs are extended more in longer hops, as is suggested by the Azizi and Abbott (2013) strain data, this could be the result of changing the rate of limb joint extension before impact and/or changing the duration over which these movements occur. These possibilities suggest distinct motor strategies that vary in complexity to achieve the same result at impact.

In this study, we examined the 3D kinematics of the elbow joint and humerus in hopping cane toads to test whether elbow and humeral kinematics vary with hop distance. In particular, we measured the elbow angle and humeral configurations throughout each hop and determined the angular excursions and durations of each phase of the hop. We hypothesized that (1) in line with muscle fascicle strain data from toads (Azizi and Abbott, 2013), forelimbs will be more extended at impact in longer hops, and (2) increased elbow extension excursions before impact will parallel increased elbow flexion excursions after impact. For any forelimb excursions that change with hop distance, we determined whether such changes are a result of alterations in velocity and/or simply the duration available for movement. As electromyographic (EMG) data suggest that elbow extensor muscles are activated later and with greater pre-landing intensity in longer hops (Gillis et al., 2010), we hypothesized that elbows begin extending later and with a greater velocity in longer hops.

## RESULTS

### Kinematics

A total of 51 hops from five animals (9–11 hops each) were used in this study. Hop distance ranged between 14 and 39 cm (mean±s.d., 25±5 cm). All other descriptive statistics are presented as means± s.d. of individual means in Tables 1 and 2 as well as in the text. Excursions of elbow extension, humeral protraction and humeral elevation are designated by positive values; elbow flexion, humeral retraction and humeral depression are designated by negative values.

### Phases of the hop

All hops could be broken down into four phases using inflection points of the elbow angle, α (Figs 1, 2). The first phase, hop initiation, begins with the onset of animal movement (when the velocity of the toad increases beyond 5 cm $s^{-1}$) and ends when the elbow begins to flex ($\alpha_1$ at $T_1$; Fig. 2C). The second phase, forelimb lift-off, lasts until the elbow stops flexing ($\alpha_2$ at $T_2$; Fig. 2C). The third phase, impact preparation, involves extension of the elbow and lasts until impact, at which point the elbow begins to flex again, signaling the final, landing phase ($\alpha_3$ at $T_3$; Fig. 2C). The landing phase ends when the elbow stops flexing after impact ($\alpha_4$ at $T_4$; Fig. 2C). Despite these stereotypical phases, there was considerable variation in the forelimb kinematics measured both within and between individuals (Fig. 3).

### Hop initiation

Animals begin hops in a resting position with their elbows at close to a right angle ($\alpha_0$=79±14 deg) and humeri pointed posteriolaterally ($\beta_0$=49±17 deg), and slightly toward the ground ($\delta_0$=172±11 deg) (see Fig. 1 for definitions of α, β and δ; $\alpha_0$, $\beta_0$ and $\delta_0$ represent angular values at time 0, e.g. Fig. 2). Hop initiation begins as the hindlimbs start to extend and the animal is pushed up

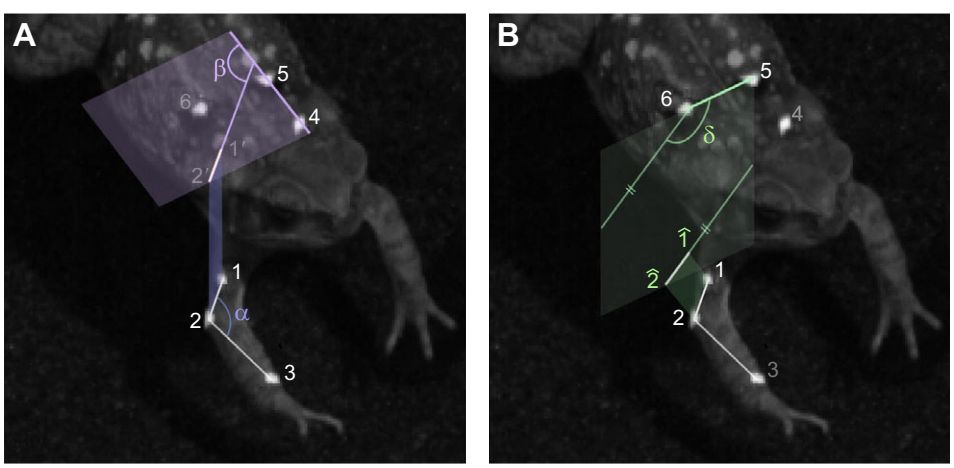


**Fig. 1. Markers and their use in calculating elbow and humeral kinematics.** White points and numbers designate marker positions on the toads. Line segments are referred to by their end point marker numbers such that the line segment between points 1 and 2 will be segment $\overline{1\,2}$ and a line through those points is $\overleftrightarrow{1\,2}$. Elbow extension/flexion angle, α, was defined as the angle between the segments $\overline{1\,2}$ and $\overline{2\,3}$ in 3D space (A). Humeral position was broken down into projections of the humeral segment $\overline{1\,2}$ onto the horizontal and vertical planes. Humeral protraction/retraction angle, β, was calculated by projecting the humeral segment $\overline{1\,2}$ onto the horizontal plane as $\overline{1'\,2'}$ (A). Protraction/retraction was calculated as the angle between the line $\overleftrightarrow{1'\,2'}$ and the central axis of the body, defined by the line $\overleftrightarrow{4\,5}$. When the humerus was perpendicular to the central axis of the body, humeral protraction/retraction angle was 90 deg. Humeral elevation/depression angle, δ, was calculated by projecting the segment $\overline{1\,2}$ onto the vertical plane (points $\hat{1}$ and $\hat{2}$, B). Elevation/depression was defined as the angle between the line $\overleftrightarrow{5\,6}$ and $\overleftrightarrow{\hat{1}\,\hat{2}}$. Humeral elevation/depression angle was 180 deg when the humerus was parallel to the plane of the back of the toad.



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**Table 2. Mean excursion, velocity and duration for each hop phase**

| | Hop initiation | | Forelimb lift-off | | Impact preparation | | Landing | |
|---|---|---|---|---|---|---|---|---|
| Duration | 78±30 ms | | 75±16 ms | | 92±25 ms | | 57±10 ms | |
| $x$ | $x_1-x_0$ | Velocity | $x_2-x_1$ | Velocity | $x_3-x_2$ | Velocity | $x_4-x_3$ | Velocity |
| α | 10±10 | 120±125 | −29±11 | −378±127 | 47±13 | 512±100 | −38±14 | −678±201 |
| β | −12±11 | −144±119 | 27±11 | 379±165 | 45±15 | 495±122 | −55±14 | −986±204 |
| δ | 3±8 | 49±110 | −1±11 | 5±165 | −37±10 | −424±106 | 29±10 | 539±139 |

Limb excursion ($x_1-x_0$, etc.) is in deg and velocity is in deg $ms^{-1}$.

and forward (Fig. 2A). During this phase there is little forelimb movement (Fig. 2), although in several animals some elbow extension ($\alpha_1-\alpha_0$=10 to 20 deg) was observed, especially when they began with their elbows in a particularly flexed configuration. The average duration of the hop initiation phase ($T_1-T_0$) was 78±30 ms.

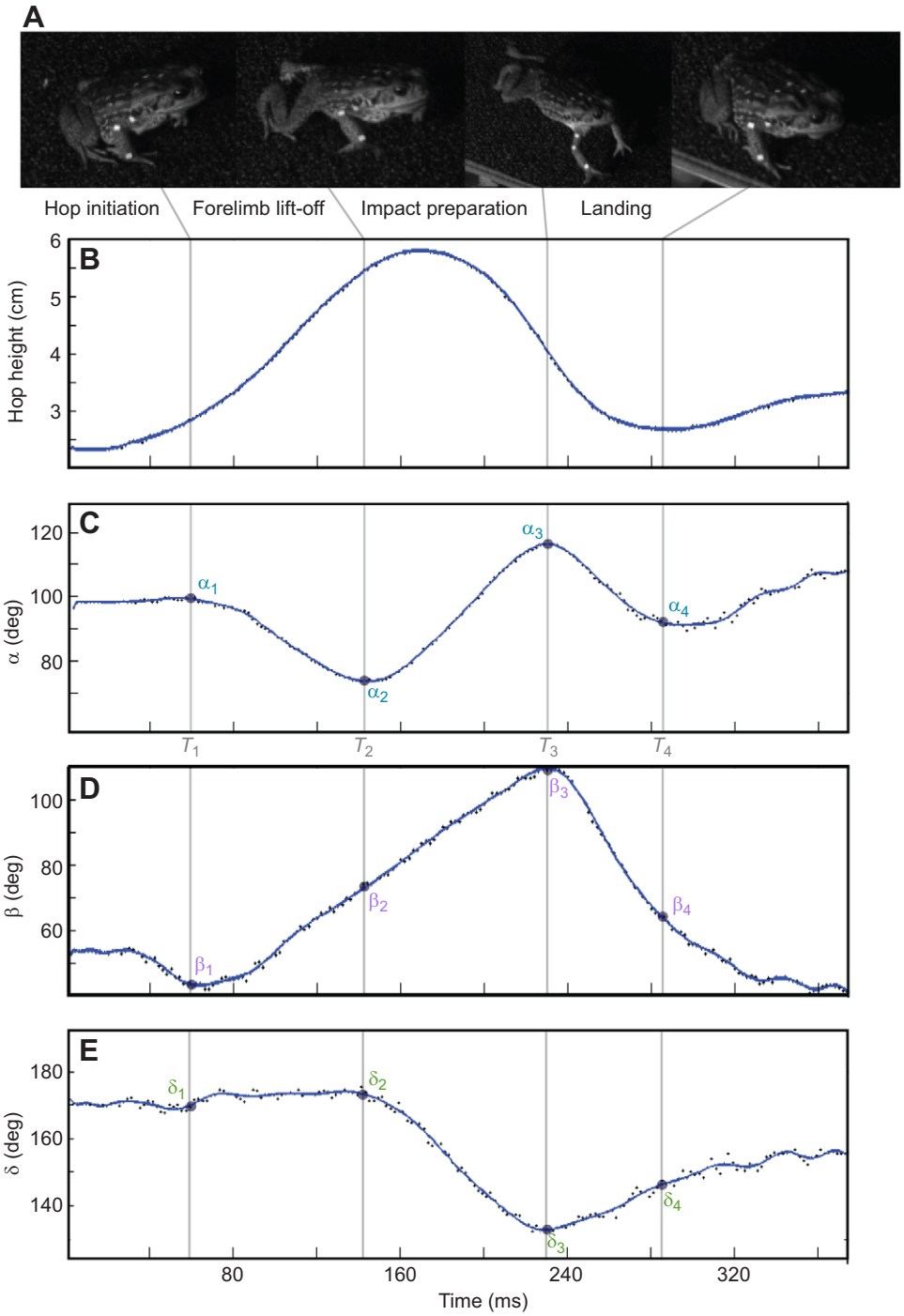


**Fig. 2. Representative kinematics from a single hop, with the four hop phases labeled and mapped onto the panels.** (A) High-speed video images representing different points in the hop. (B) Hop height. (C) Elbow extension/flexion angle α, with $T_1-T_4$ labeled on the $x$-axis representing the points used to define the beginning and end of each phase. $\alpha_1-\alpha_4$ are also indicated as the corresponding angle at those times. (D) Humeral protraction/retraction angle β, with $\beta_1-\beta_4$ indicated. (E) Humeral elevation/depression angle δ, with $\delta_1-\delta_4$ indicated. Note, traces do not return to starting values because resting postures are highly variable.

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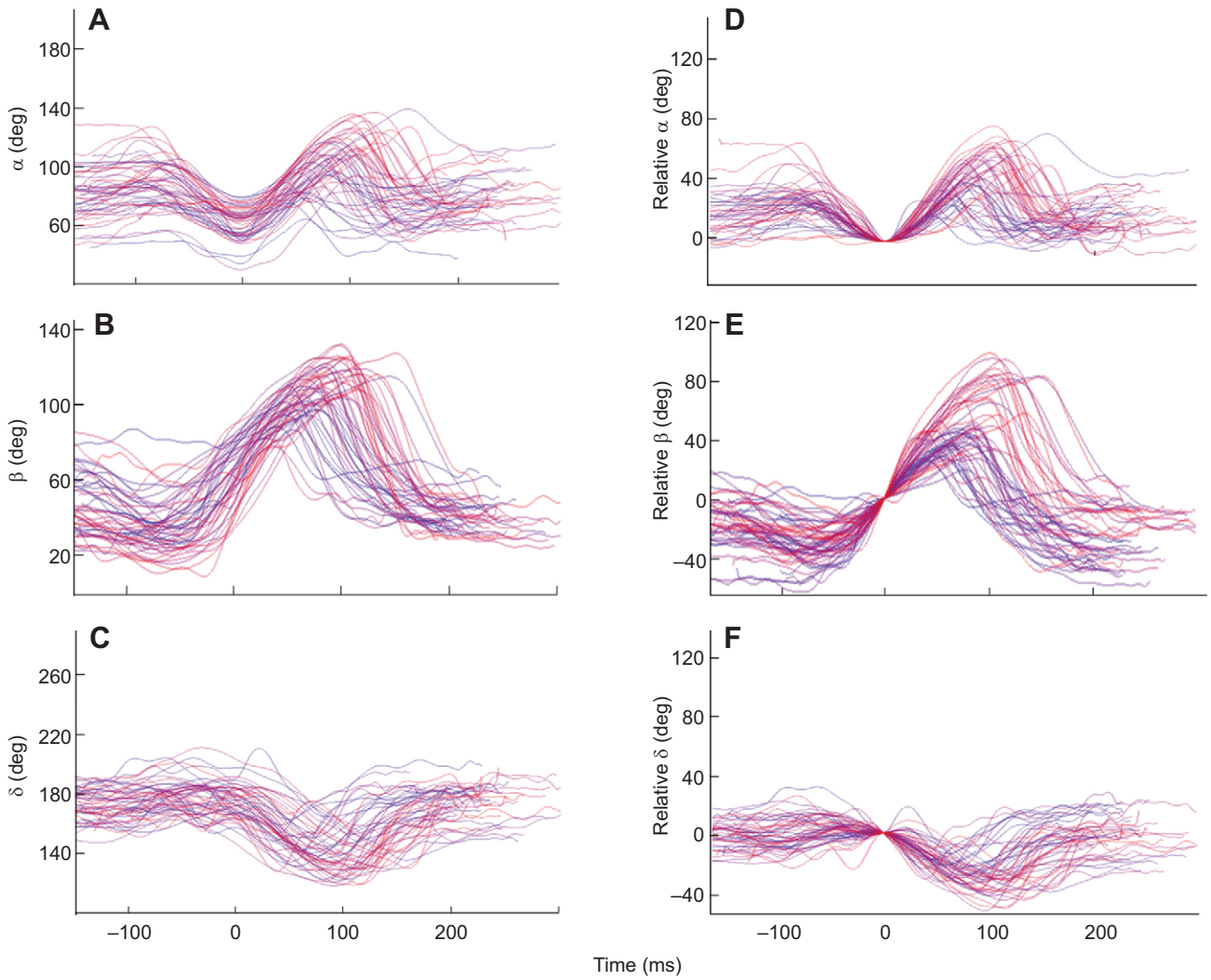


**Fig. 3. Variation in forelimb kinematics.** Elbow and humeral angles for all hops for all individuals with time zeroed at $T_2$ (A–C), and time and *y*-axis values zeroed at $T_2$ (D–F). Colors indicate hop length with warmer colors indicating longer hops.

### Forelimb lift-off

As the hindlimbs continue to extend and the toad elevates, the forelimb lift-off phase begins, and is characterized by substantial elbow flexion ($\alpha_2-\alpha_1=-29\pm11$ deg) and humeral protraction ($\beta_2-\beta_1=27\pm11$ deg) (Fig. 2). There is typically little-to-no humeral elevation or depression in this phase ($\delta_2-\delta_1=-1\pm11$ deg). The combined actions of hindlimb extension and elbow flexion lead to the forelimb losing ground contact, and humeral protraction begins to reposition the manus more anteriorly. The relative timing of this phase was highly variable in relation to hindlimb actions. For example, in some hops this entire phase occurred before hindlimb lift-off (i.e. before the animal takes off), while in others it ended much later in the aerial phase. The average duration of the forelimb lift-off phase ($T_2-T_1$) was 75±16 ms.

### Impact preparation

The impact preparation phase starts when the elbows stop flexing and begin to extend, and involves large amounts of elbow extension ($\alpha_3-\alpha_2=47\pm13$ deg), humeral protraction ($\beta_3-\beta_2=45\pm15$ deg) and humeral depression ($\delta_3-\delta_2=-37\pm10$ deg) (Fig. 2). All of these movements serve to position the manus more anteriorly and ventrally (toward the ground) as the animal braces for landing. The average duration of this phase ($T_3-T_2$) was 91±25 ms and the time to the onset of impact preparation from the beginning of movement ($T_2-T_0$) was 152±26 ms.

### Landing

At touchdown the arms are typically configured so that the elbows are extended ($\alpha_3=108\pm14$ deg) and the humeri protracted ($\beta_3=109\pm12$ deg) and depressed ($\delta_3=137\pm11$ deg) well beyond their positions at any other point in the hop (Fig. 2). Increased extension excursions at the elbow ($\alpha_3-\alpha_2$) in preparation for impact in longer hops are mirrored by increased flexion excursions after impact ($\alpha_4-\alpha_3$; Fig. 4). During the landing phase, the elbows flex ($\alpha_4-\alpha_3=-38\pm14$ deg), and the humeri are driven posteriorly ($\beta_4-\beta_3=-55\pm14$ deg) and dorsally ($\delta_4-\delta_3=29\pm10$ deg) as the body decelerates ($T_4-T_3$) over 57±10 ms. Touchdown occurred 243±24 ms after the onset of movement ($T_3-T_0$).

### Distance dependence

Forelimb kinematics are independent of distance in the first two hop phases: hop initiation and forelimb lift-off. But kinematics do vary significantly with distance during the impact preparation and landing phases. For example, while the onset of the impact

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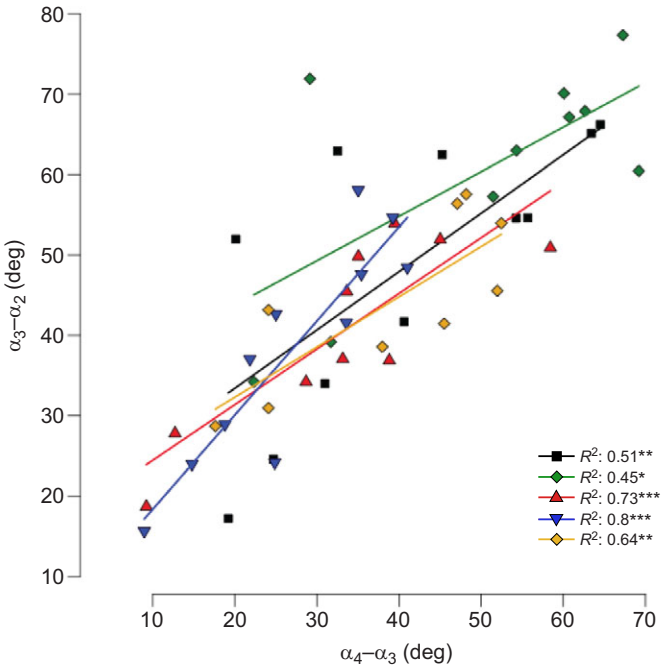


**Fig. 4. Elbow extension excursions during impact preparation, $\alpha_3-\alpha_2$, versus elbow flexion excursions during landing, $\alpha_4-\alpha_3$.** Different symbols and colors represent different animals, and regression lines reflect significant relationships for individual toads. Asterisks refer to the significance level of the *P*-value for the fit. **P*<0.05, ***P*<0.01, ****P*<0.001.

preparation phase did not occur later with distance (*P*=0.47; Table 3), the duration of this phase increased with longer hops (*P*<0.001; Table 3), as the animal remained in the air longer before impact. Yet, neither elbow extension nor humeral protraction velocity changed significantly with hop distance during this phase, and as a result, this increased duration led to significantly greater elbow excursions ($\alpha_3-\alpha_2$; Fig. 5A) and humeral protractions ($\beta_3-\beta_2$; Fig. 5B) during longer hops (*P*<0.001 for both cases; Table 3), and to a more extended ($\alpha_3$; Fig. 5C) and protracted ($\beta_3$; Fig. 5D) forelimb configuration at impact (*P*<0.001 for both cases) (Table 3). Humeral depression excursions during impact preparation ($\delta_3-\delta_2$) also increased significantly with hop length (*P*<0.001); however, these increased excursions did not result in significantly different humeral depressions at impact ($\delta_3$) after Bonferroni correction (Table 3).

During the landing phase, both elbow and humeral kinematics vary with hop distance. The amount and velocity of elbow flexion ($\alpha_4-\alpha_3$), humeral retraction ($\beta_4-\beta_3$) and elevation ($\delta_4-\delta_3$) increase significantly with distance, as does the duration of the phase ($T_4-T_3$: *P*<0.001 for all cases; Table 3). Humeral elevation velocities are distance dependent during landing (Table 3). In addition, the elbow and humeral configuration at the end of the landing phase ($\alpha_4$, $\beta_4$ and $\delta_4$), when elbows are most flexed, are independent of hop length (Table 3). Thus, during landings from longer hops, the elbows start more extended and the humeri more protracted at impact but flex and retract more and faster over a longer time to end at similar configurations to those for short hops (Fig. 4, Table 3).

## DISCUSSION

Our study was motivated by the question of how toads use their forelimbs to coordinate landing hops across a range of distances. Specifically, we asked whether toad forelimbs move differently before and after landing depending on hop distance and, if so, how these different kinematic patterns are achieved. In line with our first hypothesis, we found that toad forelimbs are significantly more extended and protracted at impact in longer hops (Table 3). These more exaggerated positions are the result of greater excursions of the elbow and humerus during the impact preparation phase (Table 3, Fig. 5). In support of our second hypothesis, these distance-dependent preparatory excursions are mirrored by similarly distant-dependent amounts of elbow flexion and humeral retraction after impact (Fig. 4). However, excursions during impact preparation were not accomplished as we expected. We hypothesized that the elbows would begin to extend later in longer hops and move with a greater velocity. Instead, we found that elbow extension did not begin later in longer hops (Table 3), and extension velocities during impact preparation were independent of distance. Greater elbow excursions (and humeral protractions) were, instead, a result of the greater time available to move during longer hops (Table 3).

### Toad forelimb kinematics and control

Cane toads land in a coordinated manner and under most conditions neither the trunk nor head contacts the substrate during landing.

**Table 3. *P*-values for models fitted with each variable against hop distance in comparison to the null model**

A. Configurations

| | $T_0$ | | $T_1$ | | $T_2$ | | $T_3$ | $T_4$ |
|---|---|---|---|---|---|---|---|---|
| α | 0.49 | | 0.69 | | 0.23 | | **1.7e−10** | 0.71 |
| β | 0.90 | | 0.42 | | 0.24 | | **3.7e−07** | 0.052 |
| δ | 0.66 | | 0.81 | | 0.86 | | 5.7e−3 | 0.40 |

B. Excursions and angular velocities

| | Hop initiation | | Forelimb lift-off | | Impact preparation | | Landing | |
|---|---|---|---|---|---|---|---|---|
| Duration | 0.36 | | 0.14 | | **1.5e−5** | | **2.9e−4** | |
| Onset | 1.0 | | 0.36 | | 0.47 | | **3.8e−6** | |
| $x$ | $x_1-x_0$ | Velocity | $x_2-x_1$ | Velocity | $x_3-x_2$ | Velocity | $x_4-x_3$ | Velocity |
| α | 0.023 | 0.14 | 0.41 | 0.30 | **2.6e−11** | 2.5e−3 | **2.5e−10** | **1.7e−9** |
| β | 0.41 | 0.42 | 0.65 | 0.97 | **6.5e−7** | 0.11 | **1.4e−9** | **1.0e−6** |
| δ | 0.47 | 0.65 | 0.88 | 0.40 | **1.6e−5** | 0.98 | **5.6e−4** | 0.022 |

Hop phase duration is the length of the phase (i.e. $T_2-T_1$). Onset is the duration from the onset of movement to the onset of a phase (i.e. $T_2-T_0$).
Bold values are significant with Bonferroni correction. All significant relationships are positive correlations. When impact velocity rather than distance was used as the fixed factor, all variables showed similar significance.

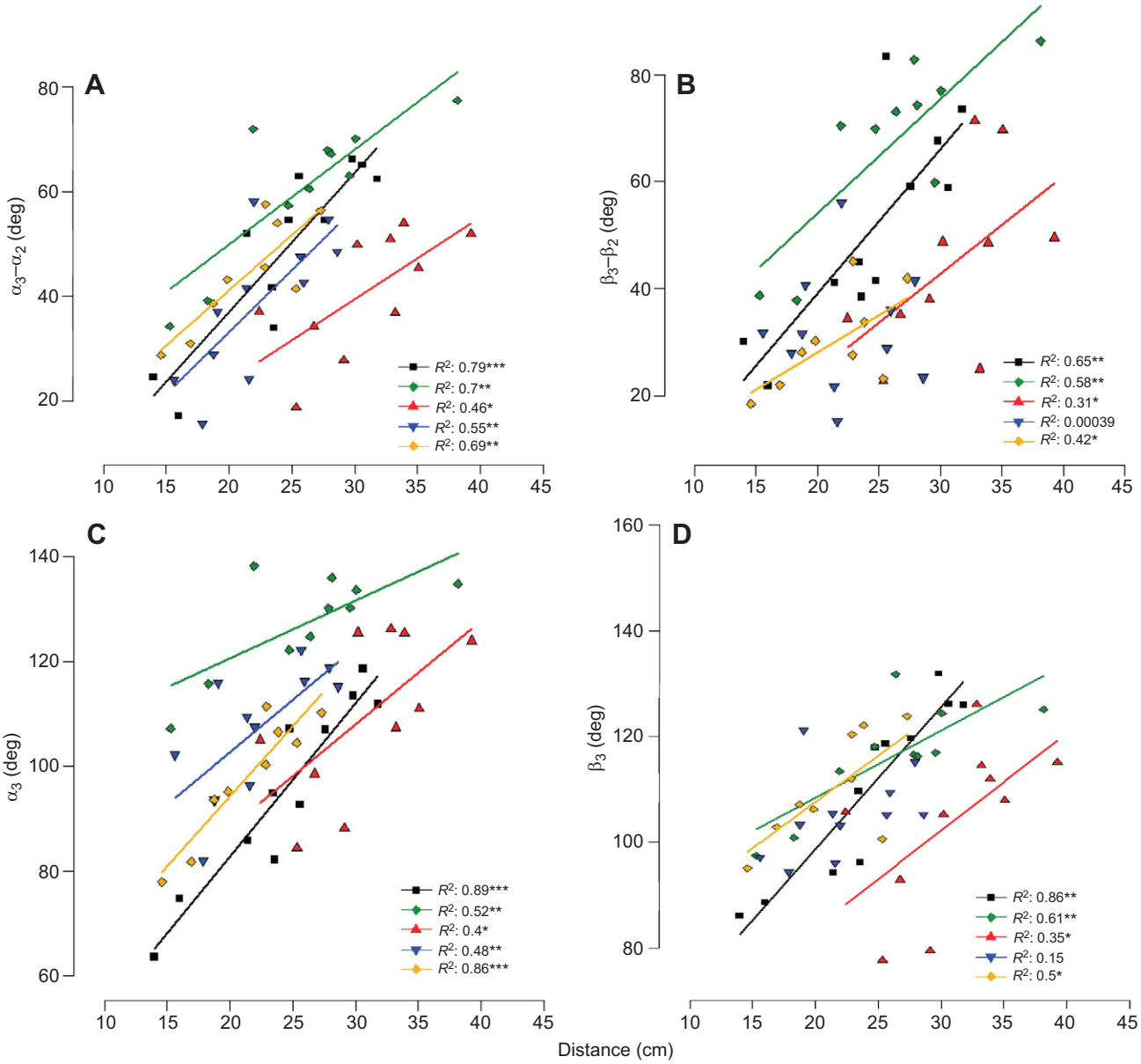


**Fig. 5. Elbow and humeral configurations and excursions regressed against distance for each animal.** (A) Elbow extension excursion ($\alpha_3-\alpha_2$) during the impact preparation phase. (B) Humeral protraction ($\beta_3-\beta_2$) during the impact preparation phase. (C) Elbow extension/flexion angle at impact ($\alpha_3$) versus hop distance. (D) Humeral protraction/retraction angle ($\beta_3$) at impact versus hop distance. For all panels, different symbols and colors represent different animals and regression lines reflect significant relationships for individual toads. Asterisks refer to the significance level of the *P*-value for the fit. **P*<0.05, ***P*<0.01, ****P*<0.001.

Instead, deceleration is exclusively controlled by the forelimbs and their underlying musculature (Azizi and Abbott, 2013; Gillis et al., 2014). Such coordination relies on appropriate pre-landing muscle activity patterns in the forelimbs (Gillis et al., 2010). However, modulating the activation timing and intensity of forelimb muscles prior to impact appears to be only one part of an integrated strategy to manage the variety of impact forces and energies associated with landing in hops of different distance. Our results indicate that cane toads also extend their elbows further during longer hops (Fig. 5A), providing a greater braking distance over which forelimb muscles can be used to decelerate the body after impact. Indeed, this is consistent with our results supporting our second hypothesis and showing that increases in preparatory elbow extension in longer hops are mirrored by subsequent increases in elbow flexion after impact (Fig. 4). As a result, the most flexed configuration of the elbow during landing does not vary with distance. Ensuring that forelimbs are more extended at impact in longer hops expands the range of impact velocities that can be managed without over-stretching muscles involved in dissipating landing energy and decelerating the body (Azizi and Abbott, 2013).

Toads modulate elbow configuration before impact using a kinematic strategy that does not change with hop distance. Toads begin to extend their elbows at roughly the same time in all hops and continue extending them at approximately the same velocity until they land. This kinematic pattern could be explained by a simple clock-like control strategy that produces forelimb landing configurations that vary predictably with distance without the need for sensory feedback. Starting elbow extension at roughly the same time in all hops allows more time for elbow extension before impact



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during longer hops simply because animals are in the air for greater durations. During shorter hops, where the landing forces are smaller, a less-extended elbow configuration is observed at impact when smaller braking distances suffice. This type of simple control strategy can even accommodate hops over terrain of variable heights. For example, animals will hit the ground later and with more force when jumping to lower landing sites, but will hit the ground with a more extended forelimb because they are in the air longer. Likewise, animals hopping up an incline will hit the ground sooner and with less force than on the level, but also with less extended forelimbs. The pattern of forelimb kinematics we observed in preparation for landing in toads is consistent with a simple control strategy that can produce functional variations in forelimb landing configurations for a range of impact velocities without the need for sensory feedback.

The strength of this strategy is also the source of its limitations, namely that it is governed by one simple rule: animals begin to extend their elbows in preparation for impact at roughly the same time and at the same rate in every hop. However, this rule implies that forelimb movements will be bilaterally symmetrical, i.e. both arms extend simultaneously. Such kinematic symmetry does not easily accommodate landings in which the toad rolls in the air or otherwise lands with one arm well before the other. Thus, a simple clock-like landing control strategy might manage landing variations related to changes in distance or height well, but may not be sufficient to accommodate asymmetrical impacts.

A more complex strategy involving the independent control of forelimbs would avoid this limitation. Moving the two forelimbs differently in anticipation of an asymmetric landing would allow for better locomotor control under more variable conditions. Yet, this level of control would require both sensory feedback to anticipate differential impact conditions of each limb and the ability to vary the timing (and/or velocity) of individual elbow excursions to brace for uneven landings. Thus, improvements in control come at a cost of more complex sensorimotor integration.

We had, in fact, hypothesized that the forelimbs would be controlled by a more complex strategy involving variation in the timing and velocity of forelimb kinematics because previous results for the elbow extensor, m. anconeus, indicated distance-dependent activation timing and intensity (Gillis et al., 2010). However, our results do not support this hypothesis. This apparent decoupling between EMG activity in an elbow extensor (m. anconeus) and the rate and timing of elbow extension may reflect the simultaneous actions of the m. coracoradialis, an antagonistic elbow flexor, which is also active throughout the impact preparation phase (Gillis et al., 2010). If this is the case, then the simple pattern of forelimb kinematics we have observed may be the result of a complex sensorimotor control strategy that also must account for antagonistic contractions important for joint stabilization at impact. Further studies that measure both kinematics and muscle activity simultaneously in non-level hops may be able to shed more light on the control strategy that toads use to perform such controlled landings.

### Comparative limb kinematics in preparation for landing

While patterns of pre-landing limb muscle activity have received a great deal of attention across a range of vertebrate jumpers (Akella and Gillis, 2011; Dyhre-Poulsen and Laursen, 1984; Magalhães and Goroso, 2009; McKinley et al., 1983; Santello, 2005; Santello and McDonagh, 1998), the details of the corresponding limb kinematics are less well known. We can infer some information about pre-landing limb kinematics from data on limb configurations in anticipation of impact. There appear to be at least two different strategies for preparing limbs for the moment of impact. In humans, as the expected force of impact increases, limbs are moved into more flexed configurations before landing (Ford et al., 2011; Hsu and Huang, 2002; Peng et al., 2011; Santello et al., 2001). Studies on the effects of knee flexion at impact show that more extended limb configurations increase maximum ground reaction force and skeletal stress while decreasing the energy absorbed by the musculature (Devita and Skelly, 1992; Louw and Grimmer, 2006; Podraza and White, 2010). In toads and turkeys, the opposite strategy is used; the landing limb is increasingly extended as the expected force of impact rises. Nevertheless, despite this increasing extension, toad and turkey limbs are far from straight at the point of impact (e.g. elbow angles in toads and knee angles in turkeys typically reach ∼140 deg in the longest hops and highest drops; Konow and Roberts, 2015), similar to knee configurations observed in landing humans (150–165 deg; Devita and Skelly, 1992; Hsu and Huang, 2002; Janssen et al., 2012). Thus, although movement patterns of limbs during impact preparation differ between species, what remains consistent is that limbs are not held fully straight at the point of impact, reducing the likelihood of hyperextension and decreasing skeletal stress while allowing muscles to dissipate much of the energy.

## MATERIALS AND METHODS

### Animals

Five adult *B. marinus* (61–124 g) were obtained from a commercial supplier and housed in groups of two to four in large plastic containers in a holding room maintained at ∼24°C with a 12 h light:12 h dark cycle. They were fed a diet of crickets several times a week and water was always available. All experimental work was approved by Mount Holyoke College's IACUC.

### Jumping trials

The toads' limbs were marked at the elbow, wrist and midway along the humerus to characterize elbow angle, and three points marking a T along the longitudinal axis of the back were additionally used to quantify humeral movements in a vertical plane (elevation/depression) and horizontal plane (protraction/retraction) (Fig. 1). Following marker placement, animals were put in a rectangular glass tank (89×43×43 cm) lined on the bottom with rough felt to ensure purchase. The tank was lit from the sides and above with two 600 W bulbs. Two high-speed cameras (Fastec HiSpec, San Diego, CA, USA) were positioned above the animal and perpendicular to each other to record simultaneous video for 3D kinematic reconstruction. Toads were placed at the end of the tank and encouraged to hop with a touch or sound. Hops were recorded at 500 frames $s^{-1}$ (1280×1024 pixels) and videos were calibrated with a 64-point 3D calibration cube.

### Data analysis

All video sequences were analyzed to identify the onset and end of animal movement for each hop. The six marked points were then digitized in each frame between these time points and 3D coordinates calculated with Matlab software (Hedrick, 2008). Data were smoothed with a quintic spline interpolation, and elbow flexion/extension angle, α, humeral protraction/retraction angle, β, and humeral elevation/depression angle, δ, were calculated as in Fig. 1. Hop distance was calculated as the horizontal distance between the starting and ending positions of a point on the back (Fig. 1). For each video frame in every hop, α, β and δ were found. Preliminary analyses of data revealed four consistent phases in each hop (described in Results), and the values of α, β and δ were identified at the start and end of each phase and used to calculate angular excursions and velocities for each phase. In addition, the duration of each phase in every hop was calculated.

### Statistics

Each of the variables, α, β and δ, at the start and end of each phase, as well as the associated excursions, velocities and durations were fitted with two

mixed linear models: a null model with no fixed effect and a full model with hop distance as a fixed effect (Bates et al., 2014). In both models, individual toads were included as random effects. The *P*-value for each full model was computed with a likelihood ratio test between the full and reduced model and corrected for multiple comparisons with a Bonferroni correction factor for the number of tests performed.

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**Competing interests**
The authors declare no competing or financial interests.

**Author contributions**
Both authors contributed to the conception, design and execution of the experiments, as well as the interpretation of the findings and drafting the manuscript.

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