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# JAVMA



# Neurological examination in healthy adult inland bearded dragons (*Pogona vitticeps*)

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#### OBJECTIVE

To evaluate neurological tests and expected results in inland bearded dragons (*Pogona vitticeps*) and generate recommendations for bearded dragon-specific neurological examination.

#### ANIMALS

26 healthy adult inland bearded dragons.

#### PROCEDURES

A complete neurological examination utilizing tests described in both mammals and reptiles was performed on each lizard, and test feasibility and outcome were recorded.

#### RESULTS

Tests with poor feasibility included oculocardiac reflex (successfully completed in 62% [16/26] of animals) and voluntary ambulation and swallowing by use of a food item (0% [0/26] of animals). Tests with outcomes considered abnormal in mammals but attributable to normal bearded dragon behavior included head position (head tilt present in 12% [3/26]) and head movement (head bob present in 4% [1/26]). Many tests had absent or inconsistent outcomes, including menace response (present in 19% [5/26]), proprioceptive positioning (present in 4% [1/26] in the thoracic limbs and 0% [0/26] in the pelvic limbs), vent reflex (present in 27% [7/26]), and myotatic reflexes (biceps present in 8% [2/26]; patellar, gastrocnemius, and triceps present in 0% [0/26]). Extensor postural thrust was absent in all successfully tested animals, but a novel reflex termed the caudal thoracic extensor reflex was noted instead in all observed animals (100% [21/21]).

#### **CLINICAL RELEVANCE**

Tests with poor feasibility or inconsistent outcomes should have low priority or be excluded from neurological examinations of inland bearded dragons. Normal behaviors should be considered for head position and movement. A bearded dragon-specific neurological examination protocol derived from these findings is described and recommended in order to decrease stress and improve neurolocalization.

**N** eurological disorders in reptiles result from infectious diseases, nutritional deficiencies, traumatic injuries, metabolic and circulatory disorders, congenital abnormalities, toxicities, and neoplasms.<sup>1-8</sup> In inland bearded dragons (*Pogona vitticeps*), specific causes of neurological disorders include, among others, *Atadenovirus* infection,<sup>9</sup> listeriosis,<sup>10</sup> hypocalcemia,<sup>11</sup> lucibufagin toxicosis from fireflies (*Photinus* sp),<sup>12</sup> encephalopathy secondary to chronic arterial hypertension,<sup>13</sup> and peripheral nerve sheath tumors.<sup>14</sup> Complete neurological examination, in combination with signalment, history, physical examination, and laboratory diagnostics, provides neuroanatomical localization and can narrow differential diagnoses as well as direct diagnostic steps, treatment, and prognosis.<sup>15-17</sup>

Interpretation requires knowledge of the expected outcomes of each neurological test in a normal animal.

In most cases, reptile neurological examinations are adapted from dogs and cats and results are expected to be the same.<sup>5,17</sup> However, neuroanatomical and electrophysiological differences exist between mammals and reptiles<sup>18,19</sup> that may lead to different test outcomes. Even within mammals, expected neurological test results differ across species, such as in rabbits and chinchillas, which do not normally exhibit the menace response seen in dogs and cats.<sup>20,21</sup> Species-specific knowledge of the expected test outcomes should be applied whenever possible. In addition, some reptile medicine references describe neurological reflex or response tests that are not commonly performed in mammals, such as the oculocardiac reflex and the righting reflex.<sup>17,19,22</sup> While there are anecdotal reports of the expected results of these tests in reptiles, these have yet to be evaluated systematically.

Normal neurological examination results have been described in 5 healthy sea turtles.<sup>5</sup> Some tests could not be performed due to the aquatic nature and limb conformation of the animals, including gait analysis on land, wheelbarrowing, hopping, and tests for conscious proprioception.<sup>5</sup> The examination protocol has been refined for use in sea turtles sustaining trauma.<sup>16</sup> Normal neurological examination findings have not been comprehensively characterized in other reptile species, including the inland bearded dragon.

Species-specific characterization of normal neurological examination findings can prevent misinterpretation and improve diagnostic utility; additionally, examination time and patient stress can be reduced by eliminating tests of low diagnostic value and prioritizing others.<sup>20,21</sup> The aims of this study were to evaluate neurological tests and expected results in apparently healthy inland bearded dragons and generate recommendations for a bearded dragon-specific neurological examination.

# **Materials and Methods**

#### Animals

Twenty-six 1- to 2-year-old intact captive-born inland bearded dragons were evaluated at a commercial breeding facility. Complete physical examinations were performed on each animal prior to enrollment in the study. Animals were excluded if they exhibited signs of distress or significant physical examination abnormalities that could have impeded complete neurological examination. Preestablished exclusion criteria included depressed or otherwise inappropriate mental status, absence of more than one-third of the tail, absence of a limb, absence of multiple digits on 1 foot, decreased range of motion in appendicular joints, spinal deformity, ocular abnormalities, and active wounds. Minor abnormalities such as dysecdysis, small skin lesions, a single missing digit per foot, or absence of only the most distal tail were considered acceptable.

All animals were adequately housed, with ambient room temperature maintained between 26.4 to 30 °C during the day (05:00 to 17:00). Within each cage, individual heat lamps provided a range up to 33.3 °C. Light from 5% UVB fluorescent bulbs was provided from 05:00 to 17:00. Night temperatures were 26.7 to 28.9 °C. All bearded dragons in this study were outside their brumation period. An appropriate diet, including insects, greens, calcium, and vitamin supplementation, was provided daily. Body weights ranged from 320 to 600 g (median, 390 g; mean, 400.4 g). The animals remained in the animal rooms at the breeding facility during the study and were returned to their enclosures following neurological examination. This study was approved by the University of California-Davis Institutional Animal Care and Use Committee under protocol No. 21467.

#### **Neurological examination**

Neurological examinations were performed by a veterinary neurologist (VDM) with restraint by a sin-

gle handler (DKT). Materials used for the examination included a nonslip mat placed on an examination table, a foam kneeling pad, paper notecards, ultrasonic doppler flow detector, ultrasound transmission gel, transilluminator, cotton-tipped applicators (CTAs), reflex hammer, hemostatic forceps, cotton balls, and a syringe with a catheter attached (6-mL Luer lock syringe; 0.7 X 19-mm [24-gauge 0.75-inch] catheter without stylet). Ambient temperature was measured with a digital infrared thermometer prior to every examination to ensure it remained within the preferred optimal temperature zone for bearded dragons.<sup>23</sup>

The neurological examination was performed as previously described for reptiles<sup>1,5,17,18</sup> or as described for mammals if a reptile-specific recommendation was not available, taking care to evaluate for consistently repeatable behavioral responses.<sup>22</sup> It consisted of 4 sections including initial observations, cranial nerve evaluation, postural reactions, and spinal reflexes.

If signs of stress developed during examination, such as rapid limb movement, tail whipping, beard or coelom inflation, change to black skin coloration, or biting attempts, the examination was paused for 1 to 2 minutes until the animal appeared calm. If an animal remained agitated despite a break or repeatedly developed signs of stress, the examination was terminated and the animal was returned to its enclosure; data collected up to the point of dismissal were included for these animals.

#### Initial observations

Initial observations were recorded in the enclosure and immediately upon placement on the mat and included mental status, body position, head position, head movement, and ambulation. Potential characterizations for head position included lowered, raised, tilted, turned, dorsoflexed, and ventroflexed. Potential abnormal head movements included tremors and bobs. Presumed high-value food items (raspberries and mealworms) were offered to encourage ambulation and swallowing. If ambulation did not occur, gentle manual pressure was applied to the pelvic limbs and tail to stimulate movement across the mat. Gait was characterized as normal, ataxic, paretic, weak, or lame, and any circling, leaning, or falling was noted. Tail tone was also noted.

#### **Cranial nerve evaluation**

Cranial nerve evaluation included testing for menace response; nictitans movement; voluntary ocular movements; resting eye position (strabismus) and nystagmus (physiological nystagmus/oculocephalic reflex and pathological nystagmus); pupil symmetry; pupillary light reflex (PLR); palpebral reflex; nasocortical stimulation response; facial sensation; corneal reflex; jaw function; masticatory muscle symmetry; facial symmetry; tongue movement, tone, and symmetry; swallowing; and oculocardiac reflex.<sup>17</sup>

The menace response was tested by use of a CTA, then again using a hand, taking care to not wave air at the subject, and the response type elicited was recorded. Nictitans movement was evaluated in conjunction with the menace response and corneal and palpebral reflexes. Resting eye position was evaluated for strabismus. Physiologic nystagmus was tested by use of 3 methods: first by manually moving just the head, then moving the body (restraining the body, leaving the head free to move), and then moving the head and body together (restraining the head and body together).<sup>22</sup> Inducible positional strabismus and nystagmus were assessed later in the examination during the righting reflex. Pupil symmetry was evaluated in ambient light. The direct and consensual PLR were assessed by means of having one evaluator watch the transilluminated eye while the other monitored for consensual response. Both the medial and lateral canthi were evaluated when the palpebral reflex was tested, as well as the upper eyelid. Nasocortical stimulation responses were assessed by use of mosquito hemostats. Facial sensation was assessed first with a CTA; if no response elicited, closed hemostats were used to apply firmer pressure. During the fifth examination, a blink reaction was seen specifically upon touching the spines at the caudal aspect of the head (parietal, postorbital, squamosal, and caudal maxillary regions; Figure 1); therefore, this specific test was added for subsequent examinations.

The corneal reflex was tested by means of blowing air from the syringe with a soft catheter tip, then touching the cornea with a dry CTA if no reflex was seen initially with air alone. Assessment of tongue movement, tone, and symmetry along with the subject's ability to swallow was evaluated by means of offering food items. If this was unsuccessful in getting the animal to open its mouth, the nose was tapped as previously described.<sup>18</sup> If still unsuccessful, an index card was used to open the mouth to assess the tongue and jaw tone. The masticatory muscles were palpated for asymmetry or atrophy and the entire face evaluated for any signs of asymmetry. The vagus nerve was assessed by means of obtaining a heart rate via a Doppler device, then rechecking the heart rate 15 seconds after applying pressure to the eyes manually with cotton balls. Although it has been recommended to apply pressure for as long as several minutes when this test is conducted,<sup>18</sup> preliminary testing showed this cohort would not tolerate a longer period.

#### **Postural reactions**

Postural reactions were evaluated on each limb by use of proprioceptive positioning (knuckling),

Figure 1-Photograph showing facial response assessment in a healthy adult inland bearded dragon (Pogona vitticeps). which was performed by means of stroking the spines overlying the parietal, postorbital, squamosal, and caudal maxillary regions with a



cotton-tipped applicator. A positive response, characterized by elevation of the lower eyelid and nictitans as seen in this image, occurred in 36% (8/22) of animals.

hopping, visual placing, and tactile placing (Supplementary Video S1). Hemiwalking, wheelbarrowing, and extensor postural thrust were also tested. Proprioceptive positioning was performed first with the body supported manually off the table and then again with the animal in a normal resting position with the tail and ventrum in contact with the table. For visual placing, the animal was elevated and slowly moved into contact with the foam kneeling pad (Figure 2); tactile placing was performed in the same manner with the eyes covered. Hemiwalking was performed on the left and right and wheelbarrowing on the thoracic limbs. Testing for extensor postural thrust was performed by means of lifting the animal horizontally off the table, then into a vertical position, slowly lowering the pelvic limbs to contact the table and then moving the animal caudally. During the fourth examination, an unexpected reaction consisting of caudal thoracic limb extension (Figure 3) was observed; thus, this was also evaluated for subsequent animals.

#### Spinal reflexes

Spinal reflexes assessed included withdrawal on all limbs and the vent, cutaneous trunci, biceps, triceps, gastrocnemius, and patellar reflexes. Withdrawal reflexes were assessed first by use of a manual digital pinch and then a light pinch with a hemo-



Figure 2—Photograph showing visual placing assessment of the left thoracic limb of a healthy adult inland bearded dragon, which was performed by use of a foam kneeling pad. Tactile placing was assessed in the same manner, with the examiner's hand covering the animal's eyes.



Figure 3—Photograph showing the caudal thoracic extensor reflex in a healthy adult inland bearded dragon, characterized by caudal extension of thoracic limbs during extensor postural thrust testing.

stat. The presence or absence of a crossed extensor reflex was noted. The vent reflex was assessed by means of light pinching with hemostats near the mucocutaneous junction of the vent in a circumferential pattern. Cutaneous trunci testing was conducted with a gentle pinch by use of hemostats starting at the distal third of the tail, extending cranially to the thoracic intumescence. Myotatic reflexes including the biceps, triceps, gastrocnemius, and patellar reflexes were tested as described for dogs and cats.<sup>22</sup> The patellar reflex was performed with the limb suspended in a non-weight-bearing position. Righting reflex testing was performed at the end of the exam, as it was deemed most likely to result in stress and behaviors impeding further examination, such as coelomic inflation. While the animal was in dorsal recumbency, if tolerated, the ventrum was stroked to test whether this would inhibit the reflex through a trancelike state as previously reported in some animals.<sup>18</sup> As the animal righted itself, it was noted whether the head or coelom led the movement.

#### **Data analysis**

Tests were considered successfully completed if the animal's behavior permitted test completion and the test could be physically performed as described in other species. A test result was considered expected if it was consistent with the described result in reptiles<sup>1,5,16-18</sup> or in mammals if no reptilian description was available.<sup>22</sup> For myotatic reflexes, the expected mammalian response<sup>22</sup> was used as the default given inconsistent results found in the sea turtle study.<sup>5</sup> In the case of tests performed bilaterally, results of the right and left side were combined. Each test result was reported as both percentages and frequencies. A 2-tailed paired t test was conducted on the heart rates measured during oculocardiac reflex testing. Tests that showed > 75% success in completion and > 75% consistent positive reaction rate were included for recommendation.

# Results

#### **Initial observations**

All but one of the initial neurological observations were completed with 100% success (**Table 1**). Offering a food item to stimulate ambulation had 0% success.

Three of 26 animals' head positions were characterized as head tilts (**Figure 4**), and 1 was characterized by vertical head bobbing. Successfully completed observations otherwise revealed expected results.

#### **Cranial nerve evaluation**

Cranial nerve tests had 100% successful completion with 2 exceptions (**Table 2**). Offering a food item to assess swallowing had 0% success. Ocular pressure application to induce an oculocardiac reflex was only tolerated in 62% (16/26) of animals. Facial response to stimulation of the spines at the caudal aspect of the head was recorded separately starting with the fifth animal.

Cranial nerve test results varied widely, with 0% to 100% of animals demonstrating expected results for each test. A menace response was seen in only 19% (5/26) of animals; 4 animals blinked, and 1 moved its head. No menace responses were produced by use of a moving CTA; all 5 positive responses were generated with a hand motion. Direct PLR was seen in 77% (20/26) of animals, with the consensual reflex seen in 58% (15/26). In 6 animals, a spasm of 1 or both irises was seen. Physiological nystagmus could be induced in all animals with all methods employed but was more pronounced if the head was immobilized with the body **(Figure 5; Supplementary Video S2)**; otherwise, the head tended to remain suspended in a stationary position despite body movement around it. Movement of



Figure 4—Photograph showing a right-sided head tilt in a healthy adult inland bearded dragon. Head tilts can be a sign of neurological disease but also occur with normal behavior, as in this case.

**Table 1**—Summary of initial observations during neurological examination of 26 healthy adult inland bearded dragons (*Pogona vitticeps*).

Test or observation	Expected result	Successful test completion	Successfully tested with expected result
Mentation	Bright and alert	100 (26/26)	100 (26/26)
Head position	Centered; raised or lowered	100 (26/26)	88 (23/26)*
Body posture	Sternal, palmigrade/plantigrade	100 (26/26)	100 (26/26)
Head movement	Static	100 (26/26)	96 (25/26)†
Ambulation for food item	Quadrupedal, symmetric gait	0 (0/26)	NA
Ambulation with manual stimulus	Quadrupedal, symmetric gait	100 (26/26)	100 (26/26)

Values represent percentage (proportion) of examined animals.

\*Three animals with head tilts were classified as abnormal in accordance with previously published reptile neurological examination criteria.<sup>1,17</sup> <sup>†</sup>One animal with head bobbing was classified as abnormal in accordance with previously published reptile neurological examination criteria.<sup>1</sup>

NA = Not applicable.

Table 2—Summary of cranial nerve testing during neurological examination of the inland bearded dragons of Table 1.

Test or observation	Expected result	Successful test completion	Successfully tested with expected resul
Menace response	Blink or aversive movement	100 (26/26)	19 (5/26)*
Pupil symmetry	Symmetric	100 (26/26)	100 (26/26)
PLR (direct)	Constriction of pupil	100 (26/26)	77 (20/26)
PLR (consensual)	Constriction of contralateral pupil	100 (26/26)	58 (15/26)
Strabismus	Absent	100 (26/26)	100 (26/26)
Physiological nystagmus	Nystagmus, fast phase in direction of head movement	100 (26/26)	100 (26/26)
Pathological nystagmus	Absent	100 (26/26)	100 (26/26)
Movement of nictitans/ lower eyelid	Present with palpebral or corneal reflex	100 (26/26)	100 (26/26)
Palpebral reflexes, medial canthus	Blink and/or aversive movement	100 (26/26)	92 (24/26)
Palpebral reflexes, lateral canthus	Blink and/or aversive movement	100 (26/26)	92 (24/26)
Upper eyelid sensation	Blink and/or aversive movement	100 (26/26)	100 (26/26)
Maxillary sensation	Blink and/or aversive movement	100 (26/26)	42 (11/26)
Mandibular sensation	Blink and/or aversive movement	100 (26/26)	23 (6/26)
Facial sensation: spines at parietal, postorbital, squamosal, and caudal maxillary regions	Blink and/or aversive movement	100 (22/22)†	36 (8/22)
Nasal sensation	Blink and/or aversive movement	100 (26/26)	35 (9/26)
Corneal reflex	Retraction of the globe	100 (26/26)	100 (26/26)‡
Jaw tone, symmetry, and position	Strong tone, symmetric, closed	100 (26/26)	100 (26/26)
Masticatory muscle atrophy	Absent	100 (26/26)	100 (26/26)
Swallowing	Symmetric pharyngeal muscle contraction	0 (0/26)	NA
Tapping nose	Open mouth	100 (26/26)	0 (0/26)
Tongue tone, symmetry, and movement	Symmetric with strong bilateral movement	100 (26/26)	100 (26/26)
Manual ocular pressure	Decrease in heart rate	62 (16/26)	44 (7/16)

\*All 5 responding animals required hand gesture; no animals reacted to cotton-tipped applicator movement. \*Not recorded for initial 4 bearded dragons. ‡All 26 animals responded to corneal contact with cotton-tipped applicator; no animals reacted to air applied with catheter and syringe.

PLR = Pupillary light reflex.

See Table 1 for remainder of key.

5-Photo-Figure graph showing physiological nystagmus (oculocephalic reflex) assessment in a healthy adult inland bearded drag-Testing was on. most reliable when the head and body were restrained and moved together as in this image.



the head alone resulted in resistance from the animals but elicited a satisfactory nystagmus.

Most animals showed palpebral reflexes with both lateral and medial stimulation (92% [24/26]).

The same 2 animals did not show a reflex with either stimulus. All animals showed a blink with upper eyelid stimulation; it was noted that due to patient size the postorbitofrontal ridge also tended to be stimulated. No corneal reflexes were seen with the catheter and syringe technique; the reflex was produced in all animals when the cornea was touched with a CTA. In all instances, the blink was characterized by movement of the lower eyelid and nictitans; no upper evelid movement was seen. Remaining facial sensation test results were variable, with < 50% of animals/test showing a response. The response to facial sensation was a blink in all responding animals, with additional aversive head movement in 4. No animals opened the mouth in response to nose tapping; 6 animals later opened voluntarily, but these appeared to be temporally unrelated defensive actions. Overall, 44% (7/16) of animals showed a decreased heart rate in response to ocular pressure. A 2-tailed paired t test conducted on the heart rates of all 16 animals successfully completing the test showed a significant (P = 0.02) difference between the initial rate and rate following ocular pressure.

#### **Postural reactions**

Most postural reactions were successfully completed. Two bearded dragons, both males, did not tolerate extensor postural thrust testing, visual placing, or tactile placing. Three bearded dragons did not tolerate ventrum stroking during righting reflex testing. Only 1 animal demonstrated proprioceptive positioning on the thoracic limbs, and no animals showed proprioceptive positioning on the pelvic limbs (Figure 6; Table 3). In many cases, no reaction

6—Photo-Figure graph showing proprioceptive positioning assessment of the left thoracic limb of a healthy adult inland bearded dragon. Foot replacement occurred in only 4% (1/26) of animals for the thoracic limbs and 0% (0/26) for the pelvic limbs.



was seen, even when the dorsal aspects of multiple feet were in contact with the mat. While 83% (20/24)of animals exhibited visual placing on the thoracic limbs, only 54% (13/24) showed tactile placing. Likewise, 83% (20/24) of animals exhibited visual placing on the pelvic limbs but only 54% (13/24) showed tactile placing. For the thoracic limbs, no animals were positive for tactile placing but not for visual placing; the same was true for the pelvic limbs. Twenty-four animals tolerated both hopping and tactile placing. One animal was negative on both tests in all limbs, and 1 animal was negative on both tests in the thoracic limbs. In 3 instances each for the thoracic and pelvic limbs, there was discordance between the 2 tests, meaning the expected result was seen on only one of the tests.

Caudal pelvic limb stepping, as seen in dogs and cats, was not observed during extensor postural thrust testing. After documentation of caudal extension of the thoracic limbs was instituted with the fourth animal, all bearded dragons successfully completing extensor postural thrust testing (100% [21/21]) were positive for this finding, which was named the caudal thoracic extensor reflex (Supplementary Video S3).

#### **Spinal reflexes**

Most spinal reflexes were assessed successfully **(Table 4)**. One animal had a significant conscious response to the hemostats during thoracic limb withdrawal testing, rendering it impossible to adequately assess reflexes. One animal (one of the males that did not tolerate all postural reactions) did not tolerate testing of the biceps, tri-

**Table 3**—Summary of postural reaction testing of the inland bearded dragons of Table 1.

Test or observation	Expected result	completion	successfully tested with expected result
Proprioceptive positioning, thoracic limbs	Replace foot in palmigrade position	100 (26/26)	4 (1/26)
Proprioceptive positioning, pelvic limbs	Replace foot in plantigrade position	100 (26/26)	0 (0/26)
Hopping, thoracic limbs	Hop and replace foot in palmigrade position	100 (26/26)	81 (21/26)*
Hopping, pelvic limbs	Hop and replace foot in plantigrade position	100 (26/26)	88 (23/26)+
Hemiwalking	Hop and replace feet in palmigrade/ plantigrade positions	100 (26/26)	96 (25/26)*
Wheelbarrowing	Ambulate with symmetric alternating thoracic limb movement	100 (26/26)	100 (26/26)*
Extensor postural thrust	Move pelvic limbs caudally upon touching floor	92 (24/26)	0 (0/24)‡
Visual placing, thoracic limbs	Place foot in palmigrade position upon contacting surface	92 (24/26)	83 (20/24)*
Visual placing, pelvic limbs	Place foot in plantigrade position upon contacting surface	92 (24/26)	83 (20/24)
Tactile placing, thoracic limbs	Place foot in palmigrade position upon contacting surface	92 (24/26)	54 (13/24)
Tactile placing, pelvic limbs	Place foot in plantigrade position upon contacting surface	92 (24/26)	54 (13/24)

\*Delayed or weak in 2 animals; counted as expected result. \*Delayed or weak in 3 animals; counted as expected result. ‡Caudal thoracic extensor reflex was detected during the fourth examination and seen in place of extensor postural thrust in all subsequent successfully completed examinations (100% [21/21] of animals).

See Table 1 for remainder of key.

Table 4—Summary of spina	al reflex testing of the inland	bearded dragons of Table 1.
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Test or observation	Expected result	Successful test completion	Successfully tested with expected result
Withdrawal, thoracic limbs	Flexion of carpus, elbow, and shoulder	96 (25/26)*	96 (24/25)
Withdrawal, pelvic limbs	Flexion of hip, stifle, and hock	100 (26/26)	96 (25/26)
Patellar	Single quick extension of stifle	100 (26/26)	0(0/26)
Gastrocnemius	Contraction of caudal thigh muscles	100 (26/26)	0(0/26)
Biceps	Contraction of biceps brachii and/or flexion of elbow	96 (25/26)	8 (2/26)
Triceps	Contraction of triceps muscle mass	96 (25/26)	0(0/26)
Crossed extensor	Absent	96 (25/26)	0 (0/25)
Cutaneous trunci	Bilateral skin twitch over thoracolumbar region	96 (25/26)	92 (23/25)
Vent reflex	Vent constriction and/or lateral tail movement	100 (26/26)	27 (7/26)
Righting reflex	Return to normal standing position	100 (26/26)	100 (26/26)
Ventrum stroking	Inhibition of righting reflex	88 (23/26)	13 (3/23)

\*In 12 animals, withdrawal reflex was absent or weak by use of finger pinch but strong with hemostats. In 1 animal, conscious response was too strong to assess withdrawal reflex.

See Table 1 for remainder of key.

ceps, crossed extensor, or cutaneous trunci reflex. Most animals exhibited withdrawal reflexes in both the thoracic and pelvic limbs (96% [24/25 and 25/26, respectively]). The same animal showed no withdrawal in either the thoracic or pelvic limbs. In 12 animals, the reflex was weak or absent with a finger pinch; hemostats were needed in many cases. No animals exhibited a crossed extensor reflex. The biceps reflex was elicited in 8% (2/26) of animals; otherwise, all myotatic reflexes were negative.

Cutaneous trunci testing resulted in a reaction in 92% (23/25) of animals; reflex distribution and appearance were inconsistent. Testing along the entire tail and coelom in each animal revealed only a localized skin twitch around the stimulated area (Supplementary Video S4), and a complete cutaneous trunci reflex was not seen in any animal. Seven animals showed skin twitching over both the tail and coelom. In 15 animals the skin twitch was only seen in the tail and in 1 animal only over the coelom. Tail flexion away from the stimulus was seen consistently with cutaneous stimulation in 4 animals, suggestive of an aversive reflex or response. Reaction to vent reflex testing was observed in 27% (7/26) of animals and was inconsistent in distribution, appearance, and repeatability. Only 1 animal showed a reflex bilaterally. On stimulation of the lateral aspect of the vent, 4 animals showed ipsilateral pelvic limb twitching or flexion and 1 showed tail flexion. Only 3 animals showed a skin twitch in proximity to the vent. Lastly, all bearded dragons (100% [26/26]) demonstrated the righting reflex. Twenty led with the head, 2 with the body, and 4 with the head and body simultaneously. Of the animals that tolerated ventrum stroking when in dorsal recumbency during this test, only 13% (3/23) showed righting reflex inhibition.

### Discussion

Most neurological tests of inland bearded dragons were completed successfully in the present

study, including many tests not feasible in sea turtles.<sup>5</sup> The 2 tests with 0% success—ambulation for a food item and swallowing—required voluntary behaviors. These behaviors may have been inhibited by human presence and potential associated stress; observer effects have been documented to affect normal behavior as well as response to noxious stimuli in lizards.<sup>24,25</sup> It is also possible that the food items selected were not of high value to this cohort. To reduce these influences, owners can be prompted to video their animals ambulating and eating at home prior to a visit or clinic cameras can be placed to record the animals without an observer present. All other tests without 100% successful completion, such as testing for oculocardiac or myotatic reflexes, required sustained physical contact. Such tests should be carefully prioritized with reference to clinical utility. Attempting such tests may result in poor compliance with the remaining examination. In 2 males, once the bearded dragon became stressed, it was difficult to complete the remainder of the examination. Exceptions might be considered if other tests evaluating the same parts of the nervous system were inconclusive or a high suspicion for disease of that neuroanatomy was raised, taking into consideration the initial assessment.

Tests with inconsistent outcomes, such as ventrum stroking during righting reflex testing, are not recommended in most cases. For these tests, it would not be possible to determine whether a lack of expected results reflected individual variation, disease, or a behavioral response to the stress of handling. Many other tests, including the menace response, proprioceptive positioning, myotatic reflexes, and vent reflex, showed a consistent lack of reaction in most individuals. For most of these tests, absent or decreased reactions are an abnormal finding in mammals but appear to be the normal result in bearded dragons. In this species, it would be impossible to determine whether such absent reactions indicated disease. Limiting the examination to tests with high diagnostic yield (Table 5), altering the exam order, and reducing examination duration will increase the chance of successfully completing the examination with minimal stress.<sup>20,21</sup> It is possible that companion beard-

#### **Table 5**—Recommended elements of a bearded dragon-specific neurological examination.

Test or observation	Expected result	Primary neuroanatomy assessed	Notes
Initial observations Mentation	Bright and alert	Ascending reticular activating system; brain stem;	-
Head position*	Centered, either raised or lowered;	Vestibular system; thalamocortex	-
Body posture	behavioral head tilt may occur Sternal, palmigrade/plantigrade	Brain stem; cerebellum; spinal column;	-
Head movement* Ambulation with manual stimulus	Static; behavioral head bobbing may occur Quadrupedal, may be bipedal; symmetric gait	Thalamocortex; cerebellum; vestibular system Spinal nerves and nerve roots; proprioception; vestibular system	- Study animals all refused to ambulate to procure food item; manual stimulation may be needed; consider asking owners to video at home; video of swallowing can be taken concurrently
Cranial nerve evaluation Pupil symmetry PLR (direct)	Symmetric Constriction of pupil; spasm/voluntary override may occur	CN II, III CN II, CN III	– Consider using assistant or mirror to simultaneously visualize contralateral pupil
Strabismus <sup>†</sup> Physiological nystagmus	Absent Nystagmus, fast phase in direction of head movement	CN III, IV, VI CN III, IV, VI, VIII	— More distinct if head is restrained together with body and the whole animal is moved
Pathological nystagmus <sup>†</sup>	Absent	CN III, IV, VI, VIII	Evaluate for resting and inducible/ positional nystagmus
Palpebral reflexes, medial canthus	Blink and/or aversive movement	CN V, ophthalmic branch; CN VII; thalamocortex if aversive movement	-
Palpebral reflexes, lateral canthus	Blink and/or aversive movement	CN V, maxillary branch; CN VII; thalamocortex if aversive movement	-
Upper eyelid sensation Corneal reflex	Blink and/or aversive movement Retraction of the globe and blink	CN V, VII CN V, ophthalmic branch; CN VI; CN VII	- Corneal contact using cotton-tipped
Jaw tone, symmetry, and position Masticatory muscle atrophy <sup>+</sup> Tongue tone, symmetry, and movement	Closed, symmetric, strong tone; behavioral gaping may occur Absent Symmetric with strong bilateral movement	CN V CN V CN XII	applicator required — Manual opening of mouth required in most cases
Postural reactions Hopping, thoracic limbs Hopping, pelvic limbs Hemiwalking Wheelbarrowing Caudal thoracic extensor reflex	Hop and replace foot in palmigrade position Hop and replace foot in plantigrade position Hop and replace feet in palmigrade/plantigrade positions Ambulate with symmetric alternating thoracic limb movement Caudal extension of thoracic limbs	Proprioception Proprioception Proprioception Proprioception Proprioception	Hop each limb individually Hop each limb individually — Test is performed in the same manner as extensor (extensor postural thrust)*
Visual placing, thoracic limbs Visual placing, pelvic limbs	Place foot in palmigrade position upon contacting surface Place foot in plantigrade position upon contacting surface	Proprioception Proprioception	
Spinal reflexes Withdrawal, thoracic limbs	Flexion of carpus, elbow, and shoulder	Thoracic intumescence	Hemostats generate stronger withdrawal reflexes than finger pinch but can elicit conscious response confounding test interpretation; recommend starting with finger pinch
Withdrawal, pelvic limbs Crossed extensor <sup>†</sup> Cutaneous trunci <sup>*</sup> Righting reflex	Flexion of hip, stifle, and hock Absent Local skin twitch and tail flexion Return to normal standing position, leading with either head or body	Lumbar intumescence Suggests chronic severe upper motor lesion Local spinal cord reflexes (suspected) Vestibular system	– – Assess for inducible positional nystagmus concurrently

Note the recommended order of examination to minimize patient stress; however, prioritization of portions of Examination must be conducted within the preferred optimal temperature zone for bearded dragons (21 to 35 G ation of portions of the neurological examination most pertinent to the individual's presentation is recommended

\*Indicates different expected result than initially hypothesized. †Indicates pathologic reflex, not evaluated by this study; thus, we are unable to recommend exclusion PLR = Pupillary light reflex. — = Not applicable.

ed dragons, such as those in a weakened condition or handled often, will be more tolerant of testing and potentially more likely to exhibit the expected responses if not stressed. It was suspected that the breeding animals used in this study were less tame than most companion bearded dragons. In compliant bearded dragons, an expanded neurological examination using tests with inconsistent completion or expected results could be considered, especially if the recommended tests were inconclusive. While absence of an expected result would not be diagnostic, presence of the expected result could indicate an intact pathway.

In some cases, test results varied from the expected result in other species but could be attributed to normal bearded dragon behavior. Three animals showed intermittent head tilts and 1 showed rapid head bobbing, both normal behaviors of the genus.<sup>26</sup> While not observed during this study, bearded dragons may exhibit other normal behaviors that appear to be unexpected compared to mammals, such as bipedal ambulation and mouth gaping.<sup>27</sup> While some of these findings, such as head tilts, can be seen with disease, it is essential for clinicians to be able to identify when they occur as normal behaviors

to avoid misdiagnoses. Findings may be normal behavior rather than disease if the movement or posture is situational such as defensive mouth gaping, there is bilateral or symmetric movement, and other neurological examination results are normal.<sup>26,27</sup> Similarly, when nystagmus was tested, if the animal was restrained by the body alone and moved, the head tended to remain in place, similar to head stabilization seen in birds.<sup>28</sup> Physiological nystagmus testing was most reliable when the head and body were restrained and moved together.

Some challenges were noted with cranial nerve evaluation. While a significant reduction in heart rate was noted during oculocardiac reflex testing and longer ocular pressure periods may have resulted in more animals demonstrating decreases in heart rate, this appeared to be stressful and increased noncompliance. Alternative vagus nerve assessments should be considered. Observation of swallowing and glottis movement has been proposed<sup>17,19</sup>; however, it is possible that these tests do not exclusively evaluate the vagus nerve without concurrently testing the glossopharyngeal and accessory nerves.<sup>29</sup> Further evaluation of bearded dragon cranial nerve anatomy is warranted. Additionally, the direct PLR was observed in most but not all animals. A stronger light, such as from an endoscope, may have resulted in higher percentages of direct and consensual PLR. While bearded dragon iridial muscles and receptors have not been evaluated, the presence of a combination of skeletal and smooth muscle is documented in crocodilians and suspected in chelonians.<sup>30,31</sup> While interspecies variation may exist and a component of autonomic control is suspected, it is evident that voluntary override is possible and care should be taken to not overinterpret iridial spasms or alterations in pupil size as hippus or autonomic dysfunction. Due to the lateral eye position in this species, it was not possible to visualize both pupils simultaneously; consensual PLR testing required a second observer. Thus, it is reasonable for a single examiner to exclude the consensual portion of this test; alternatively, a mirror could be used for simultaneous visualization of both eves.

Like many other exotic species, most bearded dragons lacked a menace response.<sup>20,21</sup> As air movement during this test can inadvertently elicit blink through corneal ophthalmic nerve branch stimulation, it is possible that even fewer than 19% of bearded dragons exhibited a true menace response.<sup>22</sup> Care was taken to minimize air current generation during the examination. However, glass placement between the examiner's hand and the subject could further reduce the risk of false positives. Lack of menace response in chinchillas and rabbits is proposed to be due to different learned responses for threat evasion or response obliteration by stress; the same mechanisms may exist in bearded dragons.<sup>20,21</sup> Despite this finding, it was still possible to evaluate the optic and facial nerves through other tests, such as direct PLR and facial response testing. A cotton ball could also be considered to assess visual tracking.<sup>17,22</sup> Most bearded dragons showed the expected blink with upper eyelid and palpebral canthi stimulation. Contact with the postorbital frontal ridge during eyelid stimulation may have affected results, but avoidance was challenging due to patient size and anatomy. Of note, it is believed that the palpebral reflex tests the trigeminal and facial nerves in reptiles<sup>17,19</sup> as in dogs<sup>22</sup>; however, in birds, the trigeminal nerve is both the afferent and efferent nerve for blink.<sup>32</sup> It is warranted to further investigate the roles of the trigeminal and facial nerves in bearded dragons. Similar to other species, a consistent facial sensation response was not seen in all facial areas.<sup>21</sup> While the cutaneous zones of the reptilian head have not been well mapped out, they were presumed to be similar to mammalian species for this assessment. In contrast to most agamids, skin receptor distribution in the closely related eastern bearded dragon (*Pogona barbata*) has high receptor density on dorsal aspects of the head, especially the labial and nasal scales.<sup>33</sup> It is possible that sensation occurred but movement did not, either because bearded dragon facial skin lacks significant movement or due to stress response or other behavior. It is also possible that reptilian epidermal mechanoreceptors and nociceptors have varying thresholds that were not reached with testing. A painful stimulus by use of a needle has been used in some examinations for sensation evaluation; although beyond the scope of our study approval, this could be considered in future investigations.<sup>1</sup> While it is unexpected to see a blink response upon touching the spines at the caudal aspect of the head, skin receptors are also

documented in the postorbital region in eastern bearded dragons.<sup>33</sup> It is suspected that this stimulation tests the mandibular branch of the trigeminal nerve, analogous to touching the ear base in the dog.<sup>22</sup> Further investigation might include cutaneous nerve mapping, as has been performed in other species, or exploring other sensation and sensory threshold testing methods.<sup>1,34,35</sup>

Some cranial nerves were not evaluated in this study. Some, such as the olfactory and glossopharyngeal, are challenging to assess in all species.<sup>17,21,22</sup> Most reptiles also have a chemosensory vomeronasal branch of the olfactory nerve as well as paired terminal nerves (cranial nerve 0), which innervate the nasal epithelium vasculature and are likely chemosensory for gonadotropin-releasing hormone; these nerves are also challenging to assess clinically.<sup>18,19</sup> History obtained from the owner about appetite as well as video of the animal eating may aid in assessment.<sup>18</sup> Direct placement of a food item or liquid into the mouth during the neurological examination could also be considered for evaluation of jaw and tongue movement but was considered likely to be stressful for this cohort and should be performed with caution in compromised animals. Auditory sensitivity has been documented in many lizard species,<sup>36</sup> but behavior-based hearing assessment is challenging and unreliable<sup>1</sup> and was not attempted in this study. Techniques such as brain stem auditory evoked response assessments are effective for hearing assessment but unlikely to be employed regularly in clinical practice.<sup>36</sup> Accessory spinal nerve assessment was not included in this study; this nerve is described as absent in some species, and it is unknown if it is present in the bearded dragon.<sup>35,37</sup> Further anatomical investigation may elucidate whether an accessory spinal nerve exists in the bearded dragon and how it might be assessed. Clinical evaluation of other specialized reptilian structures such as the pineal complex were also beyond the scope of this study but should be considered in the future.<sup>19</sup>

Proprioceptive positioning has previously been reported as useful in bearded dragons.<sup>17</sup> It is possible that stress or other behavioral response suppressed some reactions in this cohort, but this is unlikely to account for 96% of the animals failing to respond. These results were similar to those for pelvic limb proprioceptive positioning in chinchillas and rabbits, 40% and 75% of which, respectively, did not react, and supported the trend that this test is not consistently reliable in exotic species.<sup>20,21</sup> Proprioceptive positioning was tested both with the patient's weight supported off the table and with the bearded dragons in a natural resting position with the coelom and tail in contact with the table. We hypothesize that foot replacement is not essential as the limbs are minimally weight-bearing in either position. Tests with increased weight-bearing such as wheelbarrowing, hemiwalking, and visual placing more consistently produced an expected response. We recommend testing both visual placing and hopping, as some dragons demonstrated a response to one but not the other. It is unknown why visual placing produced the expected response more frequently than tactile placing. It may simply be a study design consequence, as visual placing was performed first and the animals may have fatigued or altered their behavior afterward. Alternatively, it may reflect true species variation that needs to be further investigated. Extensor postural thrust evaluation showed no

pelvic limb stepping as in mammals but instead a caudally directed thoracic limb movement (ie, the caudal thoracic extensor reflex). This was consistently seen only when the pelvic limbs made contact with the ground and not when the patient was held in the air regardless of horizontal or vertical orientation. This movement has not been reported in other species and may reflect unique neural pathways and differences in normal locomotor patterns and range of motion. We suspect the presence of ascending reflex arcs from the pelvic limbs that might mediate this thoracic limb movement toward the sensed point of contact. While we theorized this to be a reflex, cortical involvement cannot be completely ruled out and the potential pathway remains unclear. Of note, bearded dragons often rest in a similar upright position, with weight resting on their tails, making this posture more normal for them compared to other species, which may explain the presence of this movement in this species but not others.<sup>26</sup>

Many spinal reflex assessments were challenging, and while completed successfully, limitations were encountered in anatomy and technique. For example, while testing the biceps reflex, the examiner's fingers were larger than the limb, making tendon palpation impossible. The thick skin and instrument size may also have made it difficult to produce a sufficiently firm and precise stimulus. Our findings were similar to those in sea turtles, in which stretch reflexes were inconsistent or absent.<sup>5</sup> Investigation of alternative reflex hammer options may be beneficial before concluding that the reflexes do not occur. However, with the exception of the patellar reflex, most myotatic reflexes are also challenging and inconsistent in dogs and cats and often excluded from routine neurological examination.<sup>22</sup> The withdrawal reflexes gave consistent expected results and should be included; a stronger stimulus with a needle may have elicited a response from the single animal showing no withdrawal.<sup>1</sup> Cutaneous trunci reflex testing also consistently produced movement; however, the reflex elicited in this study was focally present over the tail and coelom, unlike the large reflex involving the entire dorsum reported in mammalian species.<sup>22</sup> There is conflicting information in the literature regarding the presence of this reflex in reptiles,<sup>17,18</sup> and the complete cutaneous trunci pathway as defined in mammals may not truly exist. There may be variation in cutaneous anatomy and local reflex arcs, and some tail movements may reflect local reflexes or pain responses.

The vent reflex, considered analogous to the perineal reflex, was inconsistent, and testing is not prioritized, although a needle may provide a stronger stimulus.<sup>1</sup> It is interesting that the more common result was pelvic limb twitch or flexion rather than skin movement near the vent. This movement is similar to the clasp response described in sea turtles, in which the hind flippers are brought together in response to perineal stimulation.<sup>5</sup> The neuroanatomy involved in the vent reflex is not well characterized and warrants further investigation. Reports<sup>18</sup> exist describing vent tone loss with spinal cord injuries above the level expected for cloacal innervation; this may reflect unique innervation but could also reflect that visible vent tone may be absent normally, at least in bearded dragons. The righting reflex occurred in all animals, and testing is recommended, although orthopedic causes of an abnormal result should be ruled out. While

occasionally termed a response rather than a reflex in the literature,<sup>5,38</sup> righting movement persists following decerebration in other species, consistent with a true reflex.<sup>39,40</sup> The same is likely true in lizards, which have been reported to lead with the head during the righting reflex<sup>1</sup>; while this was the predominant finding in the present study, leading with the body or the head and body simultaneously also occurred and should be considered normal in bearded dragons.

For some neurological tests, such as the crossed extensor reflex and pathological nystagmus, a positive reaction or finding is only present in abnormal animals. These tests could not be assessed with the healthy bearded dragons in this study but are typically identified when performing other recommended neurological examination sections. Future studies should apply these tests to diseased animals to evaluate validity; at this time, we recommend that the examiner look for these abnormal findings while performing the recommended neurological exam.

Animals were only handled once for examination; retesting was not attempted in animals for which examination was terminated due to stress or for the initial animals in which facial sensation at the caudal spines of the head or the caudal thoracic extensor reflex were not evaluated. While a period of rest may have allowed more data collection on expected results, this may have confounded the data as animals allowed to rest may have become more compliant or, conversely, remained stressed resulting in overriding of responses. It is possible to divide neurological examinations into multiple sessions in hospitalized animals and should be done when necessary but could prove to be challenging in the field or during outpatient examinations.

Results of this study provided guidelines for concise, clinically useful bearded dragon neurological examinations (Table 5). Additional tests with less consistent completion or expected results can be considered for compliant animals with inconclusive initial examinations, with the understanding that lack of response may be normal. Examination must be performed within the preferred optimal temperature zone for the species to avoid reduced nerve conduction or behavioral changes.<sup>18</sup> In conjunction with complete history including husbandry information, the described bearded dragonspecific neurological examination will provide direction for differential diagnoses and subsequent diagnostics and treatment.<sup>1</sup> Future investigations should work to further elucidate the neuroanatomy of this species, demonstrate the accuracy of this neurological examination in characterizing disease, and evaluate the validity of these recommendations in other lepidosaurs.

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# **Supplementary Materials**

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