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1 **A 3D MRI-based atlas of a lizard brain**

2
3 **Running title:** MRI atlas of a lizard brain

4
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27
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41 **Abstract**

42

43 Magnetic resonance imaging (MRI) is an established technique for neuroanatomical
44 analysis, being particularly useful in the medical sciences. However, the application of
45 MRI to evolutionary neuroscience is still in its infancy. Few magnetic resonance brain
46 atlases exist outside the standard model organisms in neuroscience and no magnetic
47 resonance atlas has been produced for any reptile brain. A detailed understanding of
48 reptilian brain anatomy is necessary to elucidate the evolutionary origin of enigmatic
49 brain structures such as the cerebral cortex. Here, we present a magnetic resonance atlas
50 for the brain of a representative squamate reptile, the Australian tawny dragon
51 (*Agamidae: Ctenophorus decresii*), which has been the object of numerous ecological
52 and behavioral studies. We used a high-field 11.74T magnet, a paramagnetic contrasting-
53 enhancing agent and minimum-deformation modeling of the brains of thirteen adult male
54 individuals. From this, we created a high-resolution three-dimensional model of a lizard
55 brain . The 3D-MRI model can be freely downloaded and allows a better comprehension
56 of brain areas, nuclei, and fiber tracts, facilitating comparison with other species and
57 setting the basis for future comparative evolution imaging studies. The MRI model of a
58 tawny dragon brain (*Ctenophorus decresii*) can be viewed online and downloaded using
59 the Wiley Biolucida Server at wiley.biolucida.net.

60

61 **Keywords:** evolution, prosomeric, neuromeric, columnar, brain organization, magnetic
62 resonance imaging, reptile

63 **1 Introduction**

64

65 Squamate reptiles (lizards and snakes) comprise the second largest group of terrestrial
66 vertebrates, with more than 10,000 species (Pyrón, Burbrink, & Wiens, 2013; Reeder et
67 al., 2015; Uetz & Hošek, 2017). Due to the extent to which they occupy diverse
68 ecological niches, squamates have been recognized as an ideal group for comparative
69 studies of brain evolution and the evolution of brain-behaviour relationships (Hoops,
70 2018). For example, they are an optimal group with which to study comparative cognition
71 (Clark, Amiel, Shine, Noble, & Whiting, 2013; Leal & Powell, 2012; Northcutt, 2013)
72 and its relationship with the evolution of sociality (Whiting & While, 2017). Furthermore,
73 interest in the neurobiology of squamates is increasing, including both single-species
74 studies (Amiel, Bao, & Shine, 2017; Day, Crews, & Wilczynski, 1999; LaDage et al.,
75 2013; LaDage, Riggs, Sinervo, & Pravosudov, 2009; Lutterschmidt & Maine, 2014) and
76 comparative studies (Hoops, Ullmann, et al., 2017a; Hoops, Vidal-García, et al., 2017b;
77 Powell & Leal, 2014; Robinson, Patton, Andre, & Johnson, 2015).

78

79 Comparative studies on brain anatomy and evolution would be greatly facilitated if the
80 animal's nervous system could be rapidly visualized in an intact head, and even in live
81 specimens (Corfield, Wild, Cowan, Parsons, & Kubke, 2008). Magnetic resonance
82 imaging (MRI) is a noninvasive technique that allows for such visualization. This
83 technique is particularly useful in the case of endangered or protected species, and when
84 working with precious museum specimens that would be destroyed in the process of
85 extracting the brain. From a practical perspective, MRI is advantageous because it does
86 not require any of the labour-intensive tissue processing necessary for histology. The
87 resulting image can be viewed in any plane, allowing for brain regions and fiber tracts to
88 be viewed from multiple orientations throughout their rostral-caudal extent. In addition,
89 MRI can facilitate both inter- and intraspecific comparisons as measurements can be
90 semi-computer-automated (e.g. Lerch et al., 2008).

91

92 In order for MRI to facilitate comparative neuroscience, however, MRI atlases must be
93 available for a diversity of animal species. Such atlases are available for brains from most
94 major vertebrate lineages, including bony fishes (Kabli, Alia, Spaik, Verbeek, & De
95 Groot, 2006; Ullmann, Cowin, & Collin, 2010a; Ullmann, Cowin, Kurniawan, & Collin,
96 2010b), cartilaginous fishes (Yopak & Frank, 2009), birds (Poirier et al., 2008; Vellema,
97 Verschueren, Van Meir, & Van der Linden, 2011), and mammals (Dorr, Lerch, Spring,
98 Kabani, & Henkelman, 2008; Ullmann et al., 2012; Ullmann, Watson, Janke, Kurniawan,
99 & Reutens, 2013a; Ullmann et al., 2013b). To our knowledge, only one published study
100 has used MRI to image the brain of a reptile, the garter snake (*Thamnophis sirtalis*;
101 Anderson, Kabalka, Layne, Dyke, & Burghardt, 2000); however the resolution was not
102 sufficient to distinguish most structures. Developing an MRI atlas of a reptilian brain

103 would be the first step in conducting broad-scale comparative analyses both within
104 reptiles and across all the major vertebrate clades.

105

106 Here, we present a detailed description of the brain of an agamid lizard, the Australian
107 tawny dragon (*Ctenophorus decresii*, Duméril & Bibron, 1837; Reptilia: Agamidae),
108 using high-resolution MRI. In the atlas we identify nuclei, fiber tracts, and other
109 structures throughout the brain in coronal, sagittal, and horizontal orientations.
110 Furthermore, we describe our MRI data with reference to the neuromeric/prosomic
111 model, in addition to the traditional columnar model, since the former is more natural as
112 it relates to the fundamental divisions of the brain that are shared by all vertebrates
113 (Puelles, 2009; Puelles & Rubenstein, 2015; Puelles, Harrison, Paxinos, & Watson,
114 2013). This atlas therefore provides a new means of understanding the structure and
115 connectivity of the reptile brain.

116

117 **2 Methods**

118

119 **2.1 Specimen Acquisition**

120

121 Sixteen male tawny dragons were collected from the southern Flinders Ranges, South
122 Australia. We euthanized each lizard with an injection of 100 mg/kg sodium
123 pentobarbital and an equal volume of 2 mg/mL lignocaine. Each lizard was then
124 intracardially perfused following Hoops (2015). Magnevist was added to the fixative
125 perfusate (4% paraformaldehyde) to maximize image contrast in magnetic resonance
126 imaging (Ullmann, Cowin, & Collin, 2010a). The brains were stored at 4°C in a solution
127 of 0.1% Magnevist and 0.05% sodium azide in phosphate-buffered saline until imaging.
128 The Australian National University's Animal Experimental Ethics Committee approved
129 all research under protocol number A2011-49.

130

131 **2.2 Magnetic Resonance Imaging**

132

133 Whole-brain images of 13 tawny dragon brains (e.g. Figure 1a) were acquired using a
134 Bruker Avance 11.74 Tesla wide-bore spectrometer (Ettlingen, Germany) with a micro-
135 2.5 imaging probe capable of generating magnetic gradients of 1.50 T/m. Brains were
136 immersed in Fomblin (perfluoropolyether, Grade Y06/6, JAVAC, Sydney, Australia) and
137 placed in a 10 mm diameter Wilmad tube using a custom-built plastic holder (Hyare et
138 al., 2008). Parameters used in the scans were optimized for gray-white matter contrast in
139 the presence of Magnevist. We used a 3D fast gradient-echo sequence (FLASH; T₂*-
140 weighted), with repetition time = 40 ms, echo time = 8 ms, field-of-view = 11 × 11 × 16
141 mm, and matrix size = 110 × 110 × 160, producing an image with 50 μm³ isotropic
142 voxels.

143

144 For comparison, two brains were embedded in agarose and vibratome-sectioned at 70
145 μm . Brain sections were stained for 5 min using the DNA-binding stain SYBR-green
146 (Life Technologies Australia, Melbourne, Australia), rinsed in phosphate-buffered saline,
147 mounted in Fluoro-Gel (ProSciTech, Brisbane, Australia), and imaged using Olympus
148 fluorescence light microscopes.

149

150 **2.3 Model Generation and Analysis**

151

152 To ensure consistent measures of brain morphometry all images were first manually
153 masked such that consistent coverage of brain structures and nerve endings was achieved.
154 In the tawny dragon the olfactory bulbs are small and separated from the brain on long
155 stalks (Figure 2) and we were unable to stabilize their location in the Wilmad tube.
156 Therefore, the olfactory bulbs were included in the masked regions. The manually
157 masked areas were then set to the background value such that they were not included in
158 subsequent calculations.

159

160 Thirteen brain image datasets of $50 \mu\text{m}^3$ resolution were first re-oriented to standard
161 rostro-caudal orientation. All images were then corrected for B0 intensity inhomogeneity
162 using the N3 algorithm (Sled, Zijdenbos, & Evans, 1998). An image with a good signal to
163 noise ratio and no obvious artifacts was then manually selected from the group to create
164 an initial model by blurring. All images were then recursively matched to this evolving
165 model of average structure to create a minimum deformation average with a resulting
166 resolution of $20 \mu\text{m}^3$ (Figure 1b). The details of the model creation process can be found
167 in Janke & Ullmann (2015). The fitting stages in this case started at a resolution of 1.28
168 mm and finished with a resolution of $80 \mu\text{m}^3$. The model finished with a resolution of 20
169 μm^3 .

170

171 To compare the natural orientation of the tawny dragon brain to the orientation of our
172 model, a representative scan was acquired of a brain within a fully intact tawny dragon
173 head. The brain was automatically segmented from this scan using a combination of
174 registration to the constructed model, using the MINC toolkit (Vincent et al., 2016), and
175 manual corrections. The linear rotational component of the automatic registration was
176 used to measure the angle of alignment of the brain in the skull with respect to our model.

177

178 No description exists for the tawny dragon brain or for any agamid brain. A variety of
179 neuroanatomical references available for other lizards (including lacertids, iguanids, and
180 varanids) were used to identify brain areas, including neuroanatomical references for the
181 entire lizard brain (Del Corral, Miralles, Nicolau, Planas, & Rial, 1990; Donkelaar, 1998;
182 Medina, Marti, Artero, Fasolo, & Puelles, 1992), the telencephalon (Greenberg, 1982;

183 Northcutt, 1967; Peterson, 1981; Smeets, Hoogland, & Lohman, 1986), the diencephalon
184 (Butler & Northcutt, 1973; Cruce, 1974), the hindbrain (Cruce & Newman, 1981;
185 Donkelaar, Bangma, Barbas-Henry, Huizen, & Wolters, 2012; Schwab, 1979; Wolters,
186 Donkelaar, & Verhofstad, 1984; Wolters, Donkelaar, Steinbusch, & Verhofstad, 1985),
187 and the neuromeric domains (Díaz, Yanes, Trujillo, & Puelles, 2000; Medina, Smeets,
188 Hoogland, & Puelles, 1993; Medina, Puelles, & Smeets, 1994). Another important
189 reference is the turtle brain atlas (Powers & Reiner, 1980).

190

191 **3 Results**

192

193 The tawny dragon brain model described here can be viewed online and downloaded in
194 NIFTI format from wiley.biolucida.net. The model represents the spatial positioning and
195 intensity of each neural structure based on the non-linear averaging of thirteen tawny
196 dragon brains. Using the intrinsic three-axis nature of MRI-atlases (Ullmann, Cowin,
197 Kurniawan, & Collin, 2010b), we established a coordinate system with x-coordinates
198 running medio-laterally, y-coordinates running rostral-caudally, and z-coordinates running
199 ventro-dorsally, as per convention (Figure 3). The midline of the brain, which divides the
200 two hemispheres, has been designated as the plane $x = 0$. The center of the epiphysis
201 (defined as the y plane in which the diameter of the epiphysis reaches its maximum) has
202 been designated as the point $(x,y) = (0,0)$, following studies which use the parietal eye as
203 the point $(x,y) = (0,0)$ (Greenberg, 1982). The plane $z = 0$ is located centrally as defined
204 by the dimensions of the image: there are an equal number of z-planes above and below z
205 $= 0$. By convention, y-values increase caudally and z-values increase dorsally. The model
206 is bilaterally symmetric about the midline, therefore the positive and negative directions
207 in the x-plane are arbitrary.

208

209 Our model can be matched to novel MRIs, *in vivo* or *ex vivo*, and of different
210 preservation and scanning parameters. This process, called model-based segmentation, is
211 commonly used in medical MRI research (e.g. see Friedel, van Eede, Pipitone,
212 Chakravarty, & Lerch, 2014) and could be easily implemented in evolutionary
213 neuroscience to, for example, digitally “extract” lizard brains from the surrounding tissue
214 in an MRI. We registered our model to a representative MRI scan of an intact tawny
215 dragon head, scanned *ex-vivo*, to demonstrate the position of the brain (Figure 2a,b).
216 During the registration process, we observed that our atlas is not in the natural orientation
217 of the tawny dragon brain; it is rotated by 28° in the x-plane (Figure 2c). The tawny
218 dragon head MRI is also available for viewing and download from wiley.biolucida.net.

219

220 From our atlas, we were able to identify over 200 structures including areas, nuclei, fibre
221 tracts and ventricles (Table 1). Whenever possible, the terminology of Donkelaar (1998)
222 was used. Abbreviations follow the standard nomenclature rules as described in the brain

223 atlases co-authored by George Paxinos (e.g. Paxinos & Franklin, 2013; Paxinos &
224 Watson, 2013). Figures 4-20 show our atlas in sequential coronal sections, and we also
225 include a coronal section of the anterior spinal cord (Figure 21). Figures 22-30 show our
226 atlas in sequential horizontal sections, and figures 31-36 in sequential sagittal sections.
227 The morphology of the dragon brain in our MRI model closely matches coronal nuclear-
228 stained histological sections (Figures 37-45) and therefore our atlas is also relevant for
229 work using traditional neuroanatomical methods.

230

231 We have identified the major anatomical divisions of our atlas according to the columnar
232 (Table 2) and neuromeric (Table 3) models. The boundaries between neuromeres are
233 often seen as transverse, dark strips separating grisea. They sometimes run parallel to
234 major fiber tracts, such as the fasciculus retroflexus (adjacent to the boundary between
235 prosomeres 1 and 2), facilitating their visualization. In our model, the three prosomeres of
236 the diencephalon are clearly visible in both coronal and sagittal sections (Figure 46). The
237 divisions between the commonly used columnar regions are not easily distinguished,
238 likely because these regions are artificial; however, we have outlined them for
239 comparative purposes (Figure 46).

240

241 In typical MRI images of biological tissue, the signal intensity mainly reflects water
242 content. Since we used a T2*-weighted gradient echo, regions with higher water content
243 appear hyperintense, or close to white in shade. Regions that have low water content
244 appear hypointense, closer to black. The brain regions that have the highest water content
245 are generally regions with high concentrations of cell bodies and/or neuropil and these
246 therefore appear lightest. Fiber tracts tend to be the darkest due to extensive hydrophobic
247 myelination. Nonetheless, in all tissue types signal intensity can vary extensively due to
248 differences in cell size, extent of myelination, and neurochemistry. Signal intensity can
249 even show a gradient within a single region, for example input from the lateral forebrain
250 bundle creates an intensity gradient within the anterior dorsal ventricular ridge (Figure 7).
251 Therefore, different nuclei and fiber tracts are differentiated based not only on differences
252 in signal intensity but also by careful comparison with histological preparations and
253 published literature.

254

255 The precise localization of the ventricles is important for identifying surrounding tissue
256 regions, however these structures are particularly difficult to delineate in our atlas. Some
257 ventricles, such as the rostral part of the lateral and tectal ventricles, are filled with
258 aqueous liquid and appear white (e.g. Figures 4, 12, 13). Ventricles which have collapsed
259 during perfusion and fixation, and so do not contain any liquid, appear as thin lines of
260 slightly lighter intensity. The majority of the lateral ventricle appears this way (e.g.
261 Figure 7). The third and fourth ventricles appear black as they have filled with Fomblin,

262 the oil used to immerse the brain during imaging (e.g. Figure 16). Because of this
263 variation, we have outlined ventricles with white dashed lines.

264

265 The laminar morphology of some brain regions is readily distinguishable in our atlas,
266 particularly in the cerebral cortex, optic tectum and cerebellum. These layers are not as
267 apparent in the individual MRIs used to make the model, as the differences in intensity
268 are too weak. Only by generating the minimum deformation model from 13 MRIs is the
269 noise reduced, the contrast enhanced, and the layers easily observed. The cerebral cortex
270 generally contains three cell layers, a main or central cell layer flanked by two plexiform
271 layers. The outer and inner plexiform layers appear relatively light, while the central cell
272 layer appears either lighter or darker than the plexiform layers, depending on cortical area
273 (Figure 47; also see next paragraph). The darkest layer is the alveus that runs deep within
274 the inner plexiform layer and is continuous with the anterior and posterior pallial
275 commissures. A cell layer, the periventricular layer, exists along the surface of the lateral
276 ventricle, but is not distinguishable from the alveus in our model.

277

278 The cerebral cortex is divisible into four main areas, which are distinguishable based on
279 their relative positions and the morphology of the central cell layer. The medial cortex
280 lies above the septum and is characterized by a cell layer that is distinctly darker than the
281 surrounding plexiform layers. In the dorsomedial cortex, the cell layer widens, becomes
282 lighter in intensity and appears slightly convex. An additional cell layer, the cell plate of
283 the inner plexiform layer of the dorsomedial cortex, is visible in the inner plexiform
284 layer. The dorsal cortex shows the distinct three-layered structure with a thin, prominent
285 cell layer anteriorly that becomes less distinct as the inner plexiform layer decreases in
286 intensity posteriorly. The lateral cortex is the most indistinct because its cell layer is
287 diffuse and provides little contrast to the plexiform layers.

288

289 The reptilian optic tectum has a marked laminar organization consisting of cell layers
290 separated by fiber layers; in some reptiles a total of fourteen layers have been described
291 (Ramón y Cajal, 1891). These have been grouped in six main layers or strata (Donkelaar,
292 1998), which are readily distinguishable in our model (Figure 47). The optical layer is
293 only slightly darker than the adjacent superficial grey and fibrous layer, but the two can
294 be distinguished by the dark border between them. The central white layer is the darkest
295 layer, while the central grey layer is of intermediate intensity between the central white
296 and superficial layers. The periventricular grey layer is darker than the superficial layers
297 but lighter than the central white layer, and finally the periventricular white layer is as
298 dark as the central white layer, but much thinner (Figure 47).

299

300 It is also possible identify the three layers of the cerebellum: the outer molecular layer,
301 the central Purkinje layer, and the inner granular layer (Figure 47). In lizards, including

302 the tawny dragon, the cerebellum is everted (Donkelaar & Bangma, 1992). The Purkinje
303 layer is the darkest in our MRI, likely owing to the fact that this layer contains not only
304 the big somas of the Purkinje cells, but also a band of primarily afferent fibers. The
305 granular layer is the lightest in intensity.

306
307 Not all structures visible in our MRI model are made up of neural tissue. In the anterior
308 dorsal ventricular ridge, some arteries can clearly be seen as a series of dark spots (Figure
309 8; indicated by an asterisk). The meninges can be seen as thin, light structures around the
310 edge of the brain, particularly in images of the brain stem (e.g. Figure 17). Droplets of the
311 aqueous storage solution can get trapped around the brain when transferring them to
312 Fomblin for imaging. These appear as bright areas in some images, for example the
313 spaces between the optic tectum and the epiphysis (Figure 12) and between the optic
314 tectum and the cerebellum (Figure 14).

315

316 **4 Discussion**

317

318 **4.1 MRI as a method for studying comparative neuroanatomy**

319

320 To create an atlas with the best possible resolution, we have used a non-linear image
321 averaging strategy to create an ‘idealized’ model of a tawny dragon brain (Janke &
322 Ullmann, 2015). The model represents a significant improvement in resolution over the
323 MRIs of individual brains (Figure 1), and this technique is now a standard component of
324 the image registration process for modern structural MRI analysis (Johnson, Calabrese,
325 Badea, Paxinos, & Watson, 2012; Maldjian, Daunais, Friedman, & Whitlow, 2014;
326 Ullmann, Watson, Janke, Kurniawan, & Reutens, 2013a).

327

328 Unlike histology, in MRI brain size impacts the level of discernable detail. For example,
329 an MRI atlas of a monkey brain is able to delineate 720 structures in an image with a 0.5
330 mm³ voxel size (Maldjian, Daunais, Friedman, & Whitlow, 2014), whereas an MRI atlas
331 of a cichlid brain is able to delineate only 54 structures in an image with a 50 μm³ voxel
332 size (Simões, Teles, Oliveira, Van der Linden, & Verhoye, 2012). Though the absolute
333 voxel size in the cichlid atlas is much smaller than the voxel size in the monkey atlas,
334 voxel size relative to brain size is much smaller in the monkey atlas. This provides a two-
335 fold benefit to the monkey atlas: the larger absolute voxel size provides greater signal
336 intensity, while smaller relative voxel size provides greater spatial resolution. Together,
337 these factors allow for much more precise structural delineation in larger brains. This is
338 an important consideration for comparative neuroscience, where comparisons are often
339 made between brains that differ in size by orders of magnitude. Using multiple MRIs to
340 create a non-linear average brain model can help offset these issues in species with small
341 brains.

342

343 **4.2 The Columnar and Neuromeric Models of Brain Organization**

344

345 The study of brain structure requires a model of brain organization that sets easily
346 recognized landmarks that help identify neural structures along pre-established axes
347 (Puelles, 2009). In these models, the relative topological positions of the brain divisions
348 should be invariant, independent of differences in size and shape arising through
349 development or evolution (Nieuwenhuys & Puelles, 2015; Nieuwenhuys, Donkelaar, &
350 Nicholson, 1998). Two models are currently used to interpret brain morphology, the
351 columnar and neuromeric/prosomic models.

352

353 The columnar model of neural divisions has been the predominant model of the second
354 half of the twentieth century. It was based on the discovery of distinct functional columns
355 in the spinal cord, the alar plate or dorsal horn and the basal plate or ventral horn. The
356 model was then applied to the brain (Herrick, 1910; reviewed by Puelles, 2009). Thus,
357 the diencephalon was described as containing several dorsoventral columns, including
358 epithalamus, dorsal thalamus, ventral thalamus and hypothalamus. This description is still
359 used by many neuroscientists and is found in the majority of textbooks. However, the
360 columnar model is increasingly being recognized as unnatural because it does not
361 consider the curvature of the longitudinal brain axis and the true morphogenetic divisions
362 specified during development (reviewed by Puelles, 2009).

363

364 The neuromeric model (called the prosomic model when discussing the forebrain) was
365 employed by neuroembryologists during late nineteenth and early twentieth centuries,
366 and was based on the periodic transversal bulges (called neuromeres) in the neural tube
367 wall during embryonic development (Kupffer, 1906; Orr, 1896; Puelles, 2009; Puelles et
368 al., 2013). This model has recently experienced a resurgence due to its suitability for
369 explaining the expression patterns of developmental regulatory genes and their mutant
370 phenotypes, the results of experimental studies such as transplants and fate mappings, and
371 the trajectories of major fiber tracts (Díaz & Glover, 2002; Marín & Puelles, 1995;
372 Martínez, Marín, Nieto, & Puelles, 1995; Puelles, 2009; Puelles et al., 2013; Puelles &
373 Rubenstein, 1993; 2003; Shimamura, Hartigan, Martínez, Puelles, & Rubenstein, 1995).
374 The model is already applied in widely used brain atlases, such as the last edition of the
375 rat brain atlas (Paxinos & Watson, 2013), the Allen Developing Mouse Brain Atlas
376 (<http://developingmouse.brain-map.org/>), and the chicken brain atlas (Puelles, Martínez-
377 de-la-Torre, Paxinos, Watson, & Martinez, 2007). It is starting to be incorporated into
378 MRI atlases (Watson et al., 2017).

379

380 The neuromeric model is powerful for comparative purposes since the same
381 developmental units are found in all vertebrates (Medina, 2006; Puelles et al., 2007;

382 Puelles & Medina, 2002). For these reasons, in this study we used the neuromeric model
383 as our preferred paradigm to interpret MRI data, with the hope that this will be more
384 useful for future functional and evolutionary studies using our atlas. The boundaries
385 between neuromeres were identified as dark transversal strips (i.e. thin, cell poor areas)
386 between grisea (which appear lighter). Fiber tracts, easily followed in our 3D atlas, are
387 also useful for understanding the neuromeric organization of the tawny dragon brain, as
388 their main trajectories are often either longitudinal (i.e. parallel to the alar-basal
389 boundary) or transverse (i.e. parallel to the divisions between neuromeres). Though this
390 model is based on the natural divisions of the brain and therefore is more desirable than
391 the columnar model, the columnar model remains dominant in everyday use. Therefore,
392 we provide Table 2, Table 3, and Figure 40 comparing the major brain divisions and
393 subdivisions according to each model. The major differences occur in the forebrain, due
394 to the different interpretation of the longitudinal (rostrocaudal) axis and, consequently,
395 opposite view of the transverse (dorsoventral) divisions.

396

397 **4.3 Comparison with other squamates**

398

399 Although all lizards share a basic pattern of brain organization, there are divergences in
400 morphology that are related to the widespread morphological, ecological, and behavioral
401 differences between species. For instance, the optic tectum is larger in diurnal lizards than
402 in nocturnal ones, and the size of the cerebellum is related to the type of locomotion,
403 being smaller and simpler in limbless than quadrupedal lizards (Dacey & Sereno, 1992;
404 Donkelaar, 1998; Donkelaar & Bangma, 1992; Platel, 1976). Both the optic tectum and
405 the cerebellum of the tawny dragon are well developed, as predicted for a quadrupedal
406 diurnal lizard (Gibbons, 1979; Osborne, Umbers, & Keogh, 2013; Osborne, Umbers,
407 Backwell, & Keogh, 2012).

408

409 In the tawny dragon, we have identified the four classical cortical areas of the lizard
410 brain: the medial, dorsomedial, dorsal, and lateral cortices (Striedter, 1997). In the
411 dorsomedial cortex there is a cell plate visible in the inner plexiform layer (the CPDMCx
412 in Figure 10), close to the ventricle and is associated with a small but distinct ventricular
413 ridge and a thickening of the overlying dorsomedial cortex. A similar organization is also
414 observed in *Agama agama* (Figure 1B of Wouterlood, 1981). In other lizards, this inner
415 cell plate is not as evident, although some cell clusters can be observed in a similar
416 position (Martinez-Guijarro, Desfilis, & Lopez-Garcia, 1990; Medina et al., 1992; Smeets
417 et al., 1986). In gekkonids, lacertids, and iguanids, the cell clusters are more numerous in
418 the inner plexiform layer of the dorsal cortex instead of the dorsomedial cortex. Some of
419 these form a plate referred as the cell plate of Unger (Lacerta: Medina et al., 1992;
420 Gecko: Smeets et al., 1986) or the supraventricular layer (Iguana: Northcutt, 1967). In the

421 green anole a cell plate is visible in the medial and dorsomedial cortices; Greenberg
422 (1982) labelled it the dorsomedial interposition.
423
424 The identification of the dorsal pallium in reptiles has been controversial, for example see
425 Butler (2011) versus Puelles (2006). Based on genoarchitecture during development,
426 Desfilis *et al.* (2018) proposed that it is located in a very rostral and medial position,
427 resembling that of the avian dorsal pallium, or Wulst. This area shows a different
428 cytoarchitecture compared to medial, dorsomedial and dorsal cortices, which appear more
429 caudally (Desfilis *et al.*, 2018). In our MRI atlas, we identified this dorsal pallial area at
430 very rostral telencephalic levels and accordingly named it rostradorsal pallium (DPR;
431 Figures 4, 37a). At these very rostral levels we have also identified other pallial divisions:
432 the dorsolateral pallium (DLPR), the lateral pallium (LPR), medial pallium (LPM), and
433 the ventral pallium (including the anterior olfactory nucleus, AO).
434
435 The dorsal ventricular ridge, a structure unique to sauropsids, is likely derived from two
436 pallial divisions: most of it belongs to the ventral pallium, while its caudolateral pole
437 belongs to the ventrocaudal pallium (Desfilis *et al.*, 2018). These two sectors of the dorsal
438 ventricular ridge are evident at the caudal telencephalic levels of our atlas (e.g. Figure
439 10), since they appear as two light areas separated by a dark (i.e. cell poor) strip of tissue.
440 In *Iguana* these two areas are also separated by a cell-poor lamina (Northcutt, 1967). In
441 birds, the corresponding regions are the nidopallium and arcopallium, which are again
442 separated by a cell-poor lamina (Desfilis *et al.*, 2018).
443
444 In our model, the most prominent component of the ventrocaudal pallium is the nucleus
445 sphericus, a structure that exhibits substantial variation in size and complexity between
446 species. This nucleus is involved in vomerolfaction and receives massive afferents from
447 the accessory olfactory bulb (Lanuza & Halpern, 1997; Lohman & Smeets, 1993;
448 Martínez-García, Olucha, Teruel, Lorente, & Schwerdtfeger, 1991). The degree of
449 development of this nucleus likely relates to its chemosensory function. In species that
450 use the vomeronasal system extensively, such as snakes and lizards with forked tongues,
451 the spherical nucleus occupies a large proportion of the dorsal ventricular ridge (Cooper,
452 1995; Halpern, 1980; Schwenk, 1993). In species with a reduced vomeronasal organ, this
453 nucleus may be practically nonexistent, as is the case in *Anolis* (Greenberg, 1982). In the
454 tawny dragon, the spherical nucleus appears to be of intermediate size, similar to *Iguana*
455 (Northcutt, 1967). Both of these species, like *Anolis*, do not have forked tongues and are
456 not thought to be heavily reliant on vomeronasal input. However, unlike *Anolis*,
457 *Ctenophorus* and *Iguana* have femoral pores which produce a waxy substance (Gray,
458 1827) that is likely used for chemosensory signaling through the vomeronasal system
459 (Baeckens, Edwards, Huyghe, & Van Damme, 2015; Martin & Lopez, 2000).
460

461 **4.4 Relevance of the tawny dragon for studies on the neurobiological basis of**
462 **behavior**

463

464 Among squamate reptiles, agamids (dragon lizards, Hamilton, May, & Waters, 2015),
465 with more than 300 species, form an enormously diverse group with extensive
466 morphological, ecological, and behavioural differences between species. Agamids are
467 considered a good model for the study of evolutionary biology (Chen, Stuart-Fox, Hugall,
468 & Symonds, 2012; Melville, Ritchie, Chapple, Glor, & Schulte, 2011; Stuart-Fox &
469 Owens, 2003). In particular, the genus *Ctenophorus* has been the object of numerous
470 ecological and behavioural comparative studies (Osborne, 2005a; Stuart-Fox, Moussalli,
471 Johnston, & Owens, 2004; Umbers, Osborne, & Keogh, 2012).

472

473 Some tawny dragon populations are colour-polymorphic and each morph exhibits
474 different social and reproductive strategies (McLean, Stuart-Fox, & Moussalli, 2014;
475 Teasdale, Stevens, & Stuart-Fox, 2013; Yewers, Pryke, & Stuart-Fox, 2016). Recently,
476 there has been intense interest in studying colour polymorphic lizards as models of the
477 origin and maintenance of intraspecific phenotypic and genetic diversity (Corl, Davis,
478 Kuchta, & Sinervo, 2010; McLean & Stuart-Fox, 2014; Vercken, Massot, Sinervo, &
479 Clobert, 2007; Zamundio & Sinervo, 2003). One common finding is variation in
480 reproductive strategy between colour morphs (McLean & Stuart-Fox, 2014; Osborne,
481 2005b; 2005a; Osborne et al., 2012; Teasdale et al., 2013; Wellenreuther, Svensson, &
482 Hansson, 2014; Yewers et al., 2015; Zamundio & Sinervo, 2003). However, little
483 attention has been paid to neural differences between morphs, despite their obvious
484 potential role in driving behavioural variation, including reproductive strategies (but see
485 LaDage et al., 2009; 2013). Another *Ctenophorus* species, the painted dragon (*C. pictus*),
486 is also colour polymorphic (Healey, 2008; Olsson, Schwartz, Uller, & Healey, 2009;
487 Tobler, Healey, & Olsson, 2011), but most remaining *Ctenophorus* species are
488 monomorphic, making this genus an ideal system for studying the evolution of colour
489 polymorphism and its relationship to behavioural and neural variation. Further work
490 using colour polymorphic lizard species holds great potential in elucidating the neural
491 underpinnings of different reproductive strategies.

492

493 **5 Conclusions**

494

495 This is the first time, to our knowledge, that an MRI atlas of a lizard brain has been
496 produced. MRI is an innovative technique used frequently in the medical sciences. Here,
497 we have added the first reptile to the growing list of MRI atlases available for non-
498 traditional study organisms. The resolution obtained in this atlas is significantly higher
499 than that of other atlases for animals with similarly-sized brains. We hope this atlas
500 provides inspiration to further the study of the reptile brain, the correlation between brain

501 structure and function, and the study of brain evolution, particularly using comparative
502 methods. Only by advancing research in all these fields can we understand the general
503 principles of vertebrate brain organization and identify selective pressures and
504 mechanisms behind variation in the functional organization of the brain. We aspire to
505 develop a range of MRI atlases representing, as much as possible, the diversity of
506 vertebrates. Our goal is to make these universally available through a virtual museum,
507 similar to those provided by brain collections in traditional brick-and-mortar museums
508 (Iwaniuk, 2010; 2011), and more recently by the on-line brain collections such as the
509 Comparative Mammalian Brain Collection (<http://neurosciencelibrary.org>) and
510 BrainMaps.org (<http://brainmaps.org/>).

511

512 **7 Data Accessibility**

513

514 The MRI model of a tawny dragon brain (*Ctenophorus decresii*) and the MRI of a tawny
515 dragon head are freely available for download from the Wiley Biolucida Server at
516 wiley.biolucida.net.

517

518 **8 References**

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891

892 **Figure Legends**

893

894 **Figure 1.** A comparison between a coronal section from (a) an MRI image of a single
895 brain and (b) the MRI model of thirteen brains demonstrates that the model has far
896 superior resolution (voxel = $20 \mu\text{m}^3$) compared to the image of a single brain (voxel = 50
897 μm^3).

898

899 **Figure 2.** A three-dimensional rendering of an MRI of a tawny dragon (*Ctenophorus*
900 *decresii*) head showing the position of its brain from (a) a lateral perspective and (b) a
901 dorsal perspective. The majority of the brain (in red) is included in this model, however
902 the olfactory tracts and bulbs (in yellow) are excluded. (c) The natural position of the
903 brain inside the tawny dragon head is rotated 28° in the x-plane compared to the position
904 of our model.

905

906 **Figure 3.** Our coordinate system for the lizard brain, demonstrated using a three-
907 dimensional view of the lizard brain model with coordinate axes indicated. The bar in the
908 lower right corner = 1 mm.

909

910 **Figures 4-20.** Coronal sections through the MRI model of the tawny dragon
911 (*Ctenophorus decresii*) brain. Figures are in rostral-caudal order and each section is 25
912 voxels or $500 \mu\text{m}$ caudal to the previous section. The plane of each section according to
913 our coordinate system is indicated in the upper left corner. The bar in the lower right
914 corner = 1 mm. A list of abbreviations is found in Table 1.

915

916 **Figure 21.** A coronal section through the MRI model of the anterior spinal cord of the
917 tawny dragon (*Ctenophorus decresii*). The plane, according to our coordinate system, is
918 indicated in the upper left corner. The bar in the lower right corner = 1 mm. cc = central
919 canal, dc = dorsal column tract, DH = dorsal horn, VH = ventral horn

920

921 **Figures 22-30.** Horizontal sections through the MRI model of the tawny dragon
922 (*Ctenophorus decresii*) brain. Figures are in ventro-dorsal order and each section is 25
923 voxels or $500 \mu\text{m}$ dorsal to the previous section. The plane of each section according to
924 our coordinate system is indicated in the upper left corner. The bar in the lower right
925 corner = 1 mm. A list of abbreviations is found in Table 1.

926

927 **Figures 31-36.** Sagittal sections through the MRI model of the tawny dragon
928 (*Ctenophorus decresii*) brain. Figures are in medio-lateral order. Each section is 25
929 voxels or $500 \mu\text{m}$ lateral to the previous section except for Figure 32, which is 20 voxels
930 ($400 \mu\text{m}$) lateral to Figure 31. This is because Figure 31 is offset from the midline by 100
931 μm . The plane of each section according to our coordinate system is indicated in the

932 upper left corner. The bar in the lower right corner = 1 mm. A list of abbreviations is
933 found in Table 1.

934

935 **Figures 37-45.** Coronal histological sections (left panels) of a nuclear-stained tawny
936 dragon (*Ctenophorus decresii*) brain demonstrate similar anatomical features to
937 equivalent sections through the MRI model (right panels). Figures are in rostro-caudal
938 order. The plane of each section according to our coordinate system is indicated in the
939 upper right corner. The scale bar = 1 mm. A list of abbreviations is found in Table 1.

940

941 **Figure 46.** The divisions of the lizard brain according to different models of brain
942 organization. We have delineated the major neural subdivisions according to both the
943 neuromeric and columnar models of brain organization in (a) selected coronal sections
944 and (b) a sagittal section to demonstrate how these different models partition the brain.
945 White text labels the major rostro-caudal neural subdivisions, which are delineated by
946 broken white lines. Yellow text labels the major dorso-ventral neural subdivisions, which
947 are delineated by broken yellow lines. Black text labels commonly used minor regional
948 designations within the major subdivisions. The plane of each section according to our
949 coordinate system is indicated in the lower left corner. hp = hypothalamic prosomere, m
950 = mesomere, p = prosomere, r = rhombomere

951

952 **Figure 47.** Coronal sections through an MRI model (grey scale) and a fluorescent DNA-
953 stained brain (green) compare the appearance of (a) the medial, dorsomedial and dorsal
954 cortices, (b) the cerebellum, and (c) the optic tectum. a = alveus, CGL = central grey
955 layer, CL = cell layer, CWL = central white layer, DCx = dorsal cortex, DMCx =
956 dorsomedial cortex, EZ = ependymal zone, GL = glomerular layer, IPL = inner plexiform
957 layer, LV = lateral ventricle, MCx = medial cortex, ML = molecular layer, OL = optic
958 layer, OPL = outer plexiform layer, PGL = periventricular grey layer, PL = Purkinje
959 layer, PWL = periventricular white layer, SGFL = superficial grey and fibrous layer, TV
960 = tectal ventricle

961 **Tables**

962

963 **Table 1.** Legend of abbreviations used in figures 4-39 in alphabetical order. As per convention, nuclei, areas and other structures are
964 capitalized, while fibre tracks not (Paxinos & Franklin, 2013; Paxinos & Watson, 2013).

Abbreviation	Brain Region	Coronal Figures	Horizontal Figures	Sagittal Figures
*	Blood Vessel	8		
3n	Oculomotor Nerve	14	24, 25	31, 32
3N	Nucleus of the Oculomotor Nerve	14	25, 26	
3ND	Nucleus of the Oculomotor Nerve, dorsal part	15		
3NV	Nucleus of the Oculomotor Nerve, ventral part	15	25	32
3V	Third Ventricle	8, 9, 10, 11, 12, 13	23, 24	
4n	Trochlear Nerve	15, 16	26, 27	31, 32, 33, 34
4N	Nucleus of the Trochlear Nerve	15, 16	26	31, 32
4V	Fourth Ventricle	16, 17, 18, 19, 20	26, 27	
5d	Descending Tract of the Trigeminal Nerve	18, 19, 20		
5DM	Dorsal Motor Nucleus of the Trigeminal Nerve	17		33
5DN	Descending Nucleus of the Trigeminal Nerve	18, 19, 20	25, 26, 27	34
5me	Trigeminal Mesencephalic Tract	16		
5n	Trigeminal Nerve	16	25, 26	33, 34, 35
5Pr	Principal Nucleus of the Trigeminal Nerve	17		34
5Sp	Spinal Nucleus of the Trigeminal Nerve			32
5VM	Ventral Motor Nucleus of the Trigeminal Nerve	17	25	34
6n	Abducens Nerve	17, 18	24, 25	32
6N	Nucleus of the Abducens Nerve	17, 18	26	
7DM	Dorsal Motor Nucleus of the Facial Nerve	18		
7VM	Ventral Motor Nucleus of the Facial Nerve	18		
8n	Statoacoustic Nerve	17, 18	26, 27	32, 33, 34, 35

10DN	Dorsal Motor Nucleus of the Vagus Nerve	19, 20		
10N	Motor Nucleus of the Vagus Nerve	20		
12n	Hypoglossal Nerve	19, 20		
12N	Nucleus of the Hypoglossal Nerve	19, 20		32
a	Alveus	5, 6, 7, 8, 9, 10	27, 28, 29, 30	32, 33, 34, 35
A8	Catecholaminergic Cell Group A8	15	26	
ac	Anterior Commissure	8, 9	26	31, 32
AC	Nucleus of the Anterior Commissure	9		
Acb	Accumbens Nucleus	5, 6, 7	25, 26	31, 32
ADVR	Anterior Dorsal Ventricular Ridge	5, 6, 7, 8	27, 28, 29	32, 33, 34, 35
AHA	Anterior or Alar Hypothalamic Area	10		
AngC	Angular Cochlear Nucleus	17	27	
AO	Anterior Olfactory Nucleus	4	26	32, 33
AOT	Nucleus of the Accessory Olfactory Tract	8		
apc	Anterior Pallial Commissure	8, 9	26, 27	31
Arc	Arcuate Nucleus	11, 12, 13	22	31, 32
AS	Anterior Septal Nucleus	7, 8, 9	28	31
Au	Auricle	16	27	
BAC	Bed Nucleus of the Anterior Commissure	9	25	31
bc	Brachium Conjunctivum	16	26	34
bop	Basal Optic Tract	11, 12, 13, 14	23	
BOp	Nucleus of the Basal Optic Tract	12, 13, 14	25	34
CAq	Cerebral Aqueduct	13, 14, 15		31
CC	Central Canal of the Spinal Cord	21		
Ce	Cerebellum	13, 14, 15, 16, 17	27, 28, 29, 30	31, 32, 33, 34
CeL	Cerebellar Nucleus, lateral part	16	26	
CeM	Cerebellar Nucleus, medial	16		33
CG	Central Grey	12, 13, 14, 15, 16	25, 26, 27	31, 32
chp	Choroid Plexus	10		32
CPDMCx	Cell Plate of the Dorsomedial Cortex	10		

DB	Nucleus of the Diagonal Band	6, 7	24	32, 33
dc	Dorsal Cochlear Tract	17, 18, 19	27	32
DCx	Dorsal Cortex	5, 6, 7, 8, 9, 10	28, 29, 30	33, 34, 35
dcol	Dorsal Column Tract	20, 21		31, 32
DCoL	Nucleus of the Dorsal Column, lateral part	19, 20		32
DCoM	Nucleus of the Dorsal Column, medial part	19, 20		
DH	Dorsal Horn of the Spinal Cord	21		
DLA	Dorsolateral Amygdala	9	28	35
DLH	Dorsolateral Hypothalamic Nucleus	10, 11	24	32
DLPR	Dorsolateral Pallium, Rostral Part	4		
DLT	Dorsolateral Thalamic Nucleus	10	25, 26	
DLVe	Dorsolateral Vestibular Nucleus	17, 18	27	33, 34
DIRtF	Dorsal Nucleus of the Inferior Reticular Formation	19, 20	25, 26, 27	32, 33
DMCx	Dorsomedial Cortex	5, 6, 7, 8, 9, 10	30	32, 33, 34, 35
DMH	Dorsomedial Hypothalamic Nucleus	11		31
DMS	Dorsal Median Sulcus	13, 14, 15, 16		
DMT	Dorsomedial Thalamic Nucleus	10, 11	25, 26	31
DPR	Dorsal Pallium, Rostral Part	4		
DPT	Dorsal Pretectal Nucleus	12	25, 26	33, 34
DS	Dorsal Septal Nucleus	8	27	31
DSC	Dorsal Septal Nucleus, central part	9		
DSD	Dorsal Septal Nucleus, dorsal part	9	27	31
DSt	Dorsal Striatum	5, 6, 7, 8	26	33, 34
DTg	Dorsal Tegmental Nucleus	16		32
Ep	Epiphysis	11, 12	28, 29, 30	31
EPA	Entopeduncular Nucleus, anterior part	10		

EPP	Entopeduncular Nucleus, posterior part	13	24	
EPT	External Pretectal Nucleus	11, 12		34
EW	Edinger-Westphal Nucleus	14		
f	Fornix		25	
fi	Fimbria	10		
fr	Fasciculus Retroflexus	11, 12	25, 26	32
GL	Glomerular Layer of the Cerebellum	15		32
GP	Globus Pallidus	7		
Hb	Habenula	11	27	
hbc	Habenular Commissure	11		31
iarc	Internal Arcuate Fibres	18		
ic	Infima Commissure	19, 20		
IC	Nucleus of the Infima Commissure	20		
ICc	Intercollicular Nucleus	14, 15	27	33, 34
IMLF	Interstitial Nucleus of the Medial Longitudinal Fasciculus	13	25	
iot	Intermediate Olfactory Tract	7		
IPD	Interpeduncular Nucleus, dorsal part	14, 15	24	31
IPV	Interpeduncular Nucleus, ventral part	14, 15	23, 24	31
IR	Inferior Raphe Nucleus	17, 18, 19, 20	24, 25, 26, 27	31
IS	Inferior Septal Nucleus	8, 9		31
IsD	Isthmic Nucleus, Diffuse part	15		34
IsM	Isthmic Nucleus, Magnocellular part (pre-Isthmic or mesencephalic)	15, 16	26, 27	34
IsP	Isthmic Nucleus, Parvocellular part	16	25, 26	
LA	Lateral Amygdala	9	26, 27	36
LCx	Lateral Cortex	5, 6, 7, 8, 9	28, 29	34, 35, 36
lfb	Lateral Forebrain Bundle	5, 6, 7, 8,	24, 25, 26, 27	32, 33, 34

lfbd	Lateral Forebrain Bundle, dorsal peduncle	10, 11, 12, 13	25	
lfbv	Lateral Forebrain Bundle, ventral peduncle	10, 11, 12, 14	24, 25	34
LGD	Lateral Geniculate Nucleus, dorsal part	10	26	
LGV	Lateral Geniculate Nucleus, ventral part	10, 11	24, 25	33, 34
LHA	Lateral Hypothalamic Area	11, 12, 13	23	32, 33
LHb	Lateral Habenula	10		
LJC	Lateral Juxtacommissural Nucleus	12		
ll	Lateral Lemniscus	15, 16, 17	25	34
LL	Nucleus of the Lateral Lemniscus	15, 16, 17	24, 25	34
LLD	Nucleus of the Lateral Lemniscus, dorsal part		25, 26	34
LLV	Nucleus of the Lateral Lemniscus, ventral part			34
LoC	Locus Coeruleus	16	25, 26	33, 34
lot	Lateral Olfactory Tract	4, 5	26, 27	34, 35
LOT	Nucleus of the Lateral Olfactory Tract	5, 6, 7	26, 27	34, 35
LPO	Lateral Preoptic Area	8		32
LPR	Lateral Pallium, Rostral Part	4		
LS	Lateral Septal Nucleus	7, 8, 9	27	32
LTu	Lateral Tuberal Nucleus	12	22	
LV	Lateral Ventricle	4, 5, 6, 7, 8, 9, 10	26, 27, 28, 29	32, 33, 34, 35
lvesp	Lateral Vestibulospinal Tract	18, 20	25, 26	34
M	Mammillary Nuclei	13		31
m5n	Motor Root of the Trigeminal Nerve	17	25	
m7n	Motor Root of the Facial Nerve		25	
MA	Medial Amygdala	9	25, 26	34
MC	Magnocellular Cochlear Nucleus	18		32
MCx	Medial Cortex	5, 6, 7, 8, 9, 10, 11	28, 29, 30	31, 32, 33, 34, 35
mfb	Medial Forebrain Bundle	7, 8, 9, 10, 11, 12, 13	24, 25, 26	32

MHb	Medial Habenula	10		31
MJC	Medial Juxtacommissural Nucleus	12	26	31
ml	Medial Lemniscus	15, 16, 17, 18, 19, 20	23, 24, 25	31
ML	Molecular Layer of the Cerebellum	15		32
mlf	Medial Longitudinal Fasciculus	13, 14, 15, 16, 17, 18, 19, 20	24, 25, 26, 27	31, 32
MPC	Medial Parvocellular Nucleus	19	27	
MPO	Medial Preoptic Area	8, 9	25	31
MPR	Medial Pallium, Rostral Part		27	32, 33
MRtF	Middle Reticular Formation	17, 18	23, 24, 25	32, 33
MS	Medial Septal Nucleus	9	27	
MT	Medial Thalamic Nucleus	11	25	31
O	Oval Nucleus		26	32
oc	Optic Chiasm	8, 9, 10	22, 23	31, 32
OP	Olfactory Peduncle			31
ot	Optic Tract	9, 10, 11, 12, 13	22, 23, 24, 25, 26, 27	32, 33, 34
OT	Optic Tectum	11, 12, 13, 14, 15	25, 26, 27, 28, 29, 30	31, 32, 33, 34, 35, 36
p1Tg	p1 Tegmental Area (former Pretectal Reticular Formation, PtR)	13,	25	32, 33, 34
p3Tg	p3 Tegmental Area	12	24	32
p8n	Posterior Root of the Statoacoustic Nerve	18	26, 27	
Pa	Paraventricular Nucleus	9	24	
PaO	Paraventricular Organ	12	23	31
PaON	Paraventricular Organ Nucleus (formerly Periventricular Hypothalamic Nucleus)	11, 12	23, 24	
PB	Parabrachial Nucleus	15		

pc	Posterior Commissure	12	26, 27	31, 32
PC	Nucleus of the Posterior Commissure	11	26	32
PCN	Posterior Cochlear Nucleus	18		
PCt	Posteroventral Nucleus	11, 12		33
PDN	Posterodorsal Nucleus	12	27	32
pdt	Predorsal Tract	14, 15, 16, 17, 18, 19, 20	24, 25	31
PDVR	Posterior Dorsal Ventricular Ridge	9, 10	27, 28	32, 33
PH	Posterior or Basal Hypothalamus	12	24	
PL	Purkinje Layer of the Cerebellum	15		32
PM	Profound Mesencephalic Area	13, 14	26	34
PMN	Posteromedial Nucleus	11, 12		31, 32
ppc	Posterior Pallial Commissure	10	27	31, 32
PrPC	Principal Precommissural Nucleus	11, 12	25	32
PT	Pallial Thickening	5		
PTE	Prethalamic Eminence		25	32
PTG	Pretectal Geniculate Nucleus	11, 12	25, 26	33, 34
PVSC	Posterior Nucleus of the Ventral Supraoptic Commissure	12	24	34
R	Red Nucleus	14		32
r1Tg	r1 Tegmental Area (Reticular Isthmal Nucleus)	15	25	33
RM	Retromammillary Nucleus	13	23	31
rmc	Retromammillary Commissure	13	23	31
Rot	Rotund Nucleus	11	25	31, 32
S	Septum	5, 6		
s5n	Sensory Root of the Trigeminal Nerve	17		34
s7n	Sensory Root of the Facial Nerve	17	26	

SAT	Striatoamygdaloid Transition Area	8, 9	25, 26, 27	33, 34
SCh	Suprachiasmatic Nucleus	9	24	31
SCO	Subcommissural Organ	12	26	31
SD	Nucleus of the Supraoptic Decussation	11	23	33
sh	Septohypothalamic Tract	9, 10, 11	24, 25, 26	32
sm	Stria Medullaris	9, 10	25	32
SN	Substantia Nigra		25	33, 34
SO	Superior Olivary Nucleus	17	24	33
Sol	Nucleus of the Solitary Tract	19, 20		
sol	Solitary Tract	17, 18	26, 27	
SON	Supraoptic Nucleus	8, 9	24	32
sox	Supraoptic decussation		23	
spce	Spinocerebellar Tract	16, 17, 18	26, 27	34
Sph	Spherical Nucleus	9, 10	25, 26	35
spl	Spinal Lemniscus	15, 16, 17, 18, 19, 20	24, 25, 26, 27	32, 33, 34
SR	Superior Raphe Nucleus	15, 16	23, 24	31
SRtF	Superior Reticular Formation	15, 16	23, 24, 25	32, 33
SRtL	Superior Reticular Area, lateral part	16		
SRtM	Superior Reticular Area, medial part	16		32
STL	Bed Nucleus of the Stria Terminalis, lateral part	9		
STM	Bed Nucleus of the Stria Terminalis, medial part	9	26	33
T	Triangular Area	10		32
tbd	Tectobulbar Tract, dorsal part	13, 14	26, 27	32, 33
tbv	Tectobulbar Tract, ventral part	14	26	
tc	Tectal Commissure	12, 13	27	31, 32, 33
TG	Tectal Grey	11, 12	26, 27	33, 34

tgD	Tegmental Decussation		24	
TSC	Torus Semicircularis, central nucleus	13, 14, 15	27, 28	31, 32
TSL	Torus Semicircularis, laminar nucleus	13	27	32, 33, 34
tt	Tectothalamic Tract			31
ttc	Tect-tegmental Commissure	14		
Tu	Olfactory Tubercle	4	25	31, 32
TuL	Olfactory Tubercle, lateral part	5, 6		
TuM	Olfactory Tubercle, medial part	5, 6		
TV	Tectal Ventricle	12, 13	27, 28, 29	32, 33, 34, 35
TVe	Tangential Vestibular Nucleus	18		
TZ	Nucleus of the Trapezoid Body	18	24	32
VA	Ventral Amygdala	9		35
vece	Vestibulocerebellar Fibres	17, 18	26, 27	32
VH	Ventral Horn of the Spinal Cord	21		31
VIRtF	Ventral Nucleus of the Inferior Reticular Formation	19, 20	24, 25, 26, 27	32, 33
VL	Ventrolateral Thalamic Nucleus	10, 11	24	
VLS	Ventrolateral Septal Nucleus	8	26	32
VLVe	Ventrolateral Vestibular Nucleus	18	26	32, 33, 34
VM	Ventromedial Thalamic Nucleus	11	24	32
VMH	Ventromedial Hypothalamic Nucleus	11, 12, 13	22	31, 32
VMS	Ventromedial Septal Nucleus	7	26	31
VMVe	Ventromedial Vestibular Nucleus	18		
VP	Ventral Pallidum	7, 8		
VPT	Ventral Pretectal Nucleus	12, 13	25	
VTA	Ventral Tegmental Area	14	24	31

965

Z

Nucleus Z

12

966 **Table 2** The principal rostrocaudal and dorsoventral subdivisions of the central nervous
 967 system according to the columnar model of neural divisions. The dorsoventral
 968 subdivisions of roof and floor, which are universally present, are omitted.

Primary rostrocaudal subdivisions	Secondary rostrocaudal subdivisions	Primary dorsoventral divisions
Prosencephalon or forebrain	Telencephalon Diencephalon	Pallium Subpallium Epithalamus Dorsal thalamus Ventral thalamus Hypothalamus
Mesencephalon or midbrain	Mesencephalon or midbrain	Tectum Midbrain tegmentum
Rhombencephalon or hindbrain	Metencephalon Myelencephalon	Cerebellum Pontine tegmentum Several sensory nuclei (such as the nucleus of the solitary tract and the descending trigeminal nucleus) Medullar tegmentum
Spinal Cord	Spinal segments	Alar plate/dorsal horn Basal plate/ventral horn

969 **Table 3** The principal rostrocaudal and dorsoventral subdivisions of the central nervous
 970 system according to the neuromeric model of neural subdivisions. The dorsoventral
 971 subdivisions of roof and floor, which are universally present, are omitted.
 972

Primary rostrocaudal subdivisions	Secondary rostrocaudal subdivisions (protosegments)	Tertiary rostrocaudal subdivisions (segments)	Primary dorsoventral subdivisions	
Prosencephalon or forebrain	Secondary prosencephalon	Terminal or rostral hypothalamic prosomere (hp2) Peduncular or caudal hypothalamic prosomere (hp1)	Alar	Preoptic area Alar terminal hypothalamus
			Basal	Basal terminal hypothalamus
			Alar	Evaginated telencephalon (pallium and most subpallium) Alar peduncular hypothalamus
			Basal	Basal peduncular hypothalamus

	Diencephalon	Prosomere 3 (p3)	Alar	Prethalamie eminence Prethalamus
			Basal	p3 tegmentum
		Prosomere 2 (p2)	Alar	Epithalamus Thalamus
			Basal	p2 tegmentum
		Prosomere 1 (p1)	Alar	Pretectum
			Basal	p1 tegmentum
Mesencephalon or midbrain	Mesencephalon or midbrain	Mesomeres 1 and 2	Alar	Tectum
			Basal	Tegmentum (III motor nuclei)
Rhombencephalon or hindbrain	Prepontine (Istmo-cerebellar) division	Rhombomeres 0, 1, 2 (r0, r1, r2)	Alar	Isthmic nuclei Locus coeruleus Rostral vestibular nuclei Main trigeminal nucleus Cerebellum
			Basal	IV and V motor nuclei and nerve exits (IV at r0; V at r2)
	Pontine division	Rhombomeres 3, 4 (r3, r4)	Alar	Parts of vestibular nuclei Part of descending trigeminal nucleus
			Basal	V motor nuclei at r3, VII and VIII nerve exits at r4
	Ponto-medullary division	Rhombomeres 5, 6 (r5, r6)	Alar	Parts of vestibular nuclei Part of descending trigeminal nucleus
			Basal	VI motor nucleus and nerve exit at r5, VII motor nuclei at r6
	Medullary division	Rhombomeres 7-11 (r7-r11)	Alar	Parts of vestibular nuclei Part of descending trigeminal nucleus Nucleus of the solitary tract Nuclei of the dorsal column

			Basal	IX, X & XII motor nuclei & nerve exits
Spinal Cord	Cervical, Thoracic, Lumbar, Sacral, Coccygeal regions	Myelomeres	Alar	Dorsal Horn
			Basal	Intermediate and Ventral Horns

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