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Fire-induced reptile mortality following a management burn on Lapalala Wilderness (Limpopo Province, South Africa) with notes on the mechanisms of mortality

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Despite a well-practiced fire management paradigm within southern African conservation areas, little research has been conducted on the effect of fire on native reptile diversity (Parr and Chown, 2003; Branch, 2014) however fire-associated reptile mortality has regionally been recorded for *Cherisina angulata* (Schweigger 1812) (in Wright, 1988), *Chamaesaura anguina* (Linnaeus 1758) (in De Villiers and De Villiers, 2004; Boycott, 2015; Coombs, 2015), and *Bitis atropos* (Linnaeus 1758) (in Turner, 2014). Documenting such mortality is essential if the causal link between life history sensitivities to variations in fire regimes across habitat types is to be understood (Smith et al., 2012). Information regarding the physical mechanisms resulting in fire associated mortality is lacking for most fauna (Dickinson et al., 2010). Such information may prove useful in protecting species of conservation concern affected by modified burning practices e.g. *Acontias richardi* (Jacobbsen 1987) (in Bauer, 2014) and *Tetradactylus breyeri* (Roux 1907) (in Bates, 2014).

Here we report on the results of post-fire surveys conducted immediately after a single daytime management burn set on 26 October 2016 on Lapalala Wilderness (LW), a private game reserve in Limpopo Province, South Africa (-23.87105 S, 28.34428 E), with the objective to remove moribund herbaceous material and to stimulate vegetation rejuvenation. The site was previously burnt during 2011. The fire was started at 08:45 AM and spread using drip torches along the road network bordering the area. Level terrain with sandy soils without prominent surface rocky outcrops was selected to standardise survey sites. Each survey was conducted within an hour after burning and consisted of three observers walking 4 m-wide transects through specific burnt areas recording all reptile fatalities and collecting the specimens. Transects and thus the area surveyed varied in size (Table 1). The associated habitat where each individual was encountered was recorded as either open ground, associated with leaf litter and woody cover, or burned grass clumps. Specimens were identified to species level using Branch (1998), updating taxonomic names referencing Bates et al. (2014). External examination of specimens was conducted to record any thermal injury before dissection into 5 mm thick body sections which was stored in 10% buffered formalin. These dissected tissues were then processed and stained by haematoxylin and eosin for examination by light microscopy (Bancroft and Gamble, 2002). This process consumed all specimen material. Tissue structure was histopathologically examined, identifying diagnostic abnormalities which may suggest the cause of death for each individual.

The surveys produced 15 reptile fatalities in eight species over seven transects totalling 1,93 ha. Mean mortality density was calculated at 7.77 reptile mortalities per hectare. The fossorial skink *Acontias occidentalis* FitzSimons 1941 was the most abundant reptile collected, from five individuals followed by *Aparallactus capensis* Smith 1849 from four specimens. Two *Lycophidion capense* (Smith 1831) specimens were encountered with *Mochlus sundevallii* Smith 1849, *Gerrhosaurus flavigularus* (Wiegmann 1828), *Nucras intertexta* (Smith 1838), *Chamaeleo dilepis* Leach 1819, and *Psammophis brevirostris* Peters 1881, all represented by a single specimen.

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Table 1. Survey parameters for each post fire mortality transect.

Time of burning	Distance (m)	Total area (m²)	Observed mortalities
11:00-12:00	120	1920	4
12:00-13:00	190	760	2
13:00-14:00	130	3120	4
14:00-15:00	310	9840	5

For the total sample, 53% were collected from habitats associated with either woody plant canopies or leaf litter, 26% from open ground and 20% from burned grass clumps. Fossorial species, *A. occidentalis, A. capensis* and *M. sundevallii*, constituted 60% of the collective sample and four predominantly surface-dwelling species made up 33%. *Chamaeleo dilepis* was the only arboreal reptile species encountered with the individual found between combusted woody material. Specimens were all hot to the touch upon collection, often in contorted positions.

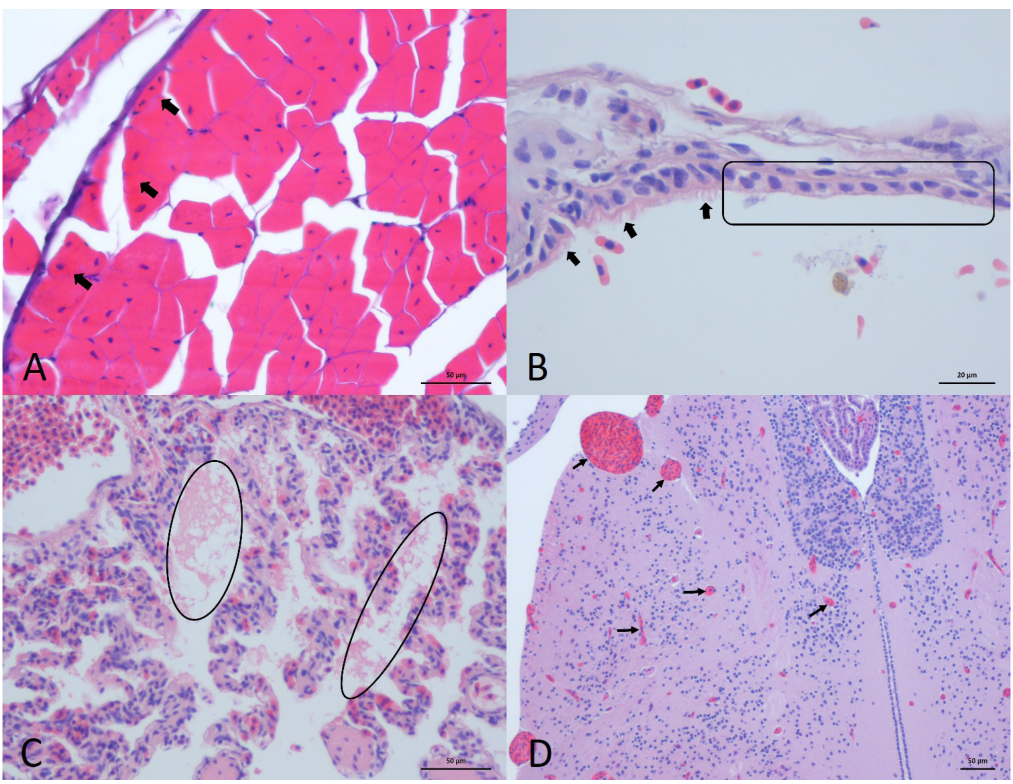

Figure 1. Histopathological manifestation of hypereosinophilic hyalinisation of peracutely thermal injured peripheral myofibres (arrows) of the outer dorso-lateral body wall (200x magnification) (A), segmental loss of tracheal epithelial ciliation (rectangle) with comparative spared ciliation of normal tracheal epithelia (arrows) of the tracheal mucosae (400x magnification) (B), amorphous hypereosinophilic fluid exudation (pulmonary oedema) in alveolar spaces (elliptical circle) in lung tissue (200x magnification) (C), and severe congestion of cerebral vasculature (arrows) in the brain (100x magnification) (D), as derived from haematoxylin & eosin stained tissue analyses. Photos by J.C.A. Steyl.

No apparent thermal damage or associated physiological responses could be detected during external examinations but one *A. capensis* specimen was desiccated and excluded from the prepared sample. Three of the collected snakes, the *P. brevirostris* and two *A. capensis*, were in advanced preparatory stages of ecdysis. Snake mortalities from fire have been associated with the preparation for ecdysis, as the dead skin likely covers and dulls selective sensory organs, making snakes slow to react to fire conditions hampering efforts to evade fire (Beaupre and Douglas, 2012).

Eleven specimens had high stomach content volumes indicating recent feeding behaviour. Evidence of increased reproductive behaviour was evident from histopathological examination. A female *M. sundevalli*i was gravid with five fully developed eggs. In addition, two *A. occidentalis*, the *P. brevirostris*, and an *A. capensis* presented various stages of egg and yolk development. Sperm was present in the testes of one *A. occidentalis* male but absent in another of an identical body length. Sperm was also observed in the epididymis of an *A. capensis* specimen with a female of the same species exhibiting sperm *in utero*. Differential seasonal timing of fires has been shown to produce varied effects on reptile populations. Spring fires are considered to produce higher mortality rates when compared to winter fires, indicating the potential importance of seasonal activity cycles (Brook & Griffiths, 2004; Beaupre and Douglas, 2011). While fossorial vertebrates are generally considered to be protected from the effects of fire (Neary et al., 1999), species-specific seasonal variations in fossoriality (e.g. Haacke and Bruton, 1978; Martin et al., 2011) may predispose certain reptiles to unfavourable conditions. The LW fire occurred during mid-spring, presumably coinciding with elevated breeding, and feeding activity in reptiles for which evidence exists in the analysed results of the mortality sample. Such heightened seasonally driven behaviours may affect the likelihood of exposure to fire conditions, either by affecting effective locomotion when carrying large volumes of ingested food or developing eggs, hindering mobility, or by stimulating surface activity, increasing the probability of fire exposure when searching for mates or during foraging activities.

An *A. capensis* and *L. capense* showed no histopathological abnormalities. A specimen of *A. occidentalis* exhibited subcutaneous muscle tissue hyalinization (Fig. 1A), with loss of cross striations and protein coagulation when compared to deeper underlying layers. Loss of respiratory epithelial ciliation (Fig. 1B) was noted in the upper respiratory tracts of all five *A. occidentalis,* single specimens of *L. capense,* and *A. capensis*, as well as the *P. brevirostris and G. flavigularus* specimens. Lung decay was observed in one

Table 2. Histopathological results per collected mortality specimen.

Specimen	Lung condition	Tracheal epithelia	Pulmonary oedema
Acontias occidentalis 1	No abnormalities	Absent	No
Acontias occidentalis 2	No abnormalities	Absent	No
Acontias occidentalis 3	Slight lung decay	Absent	No
Acontias occidentalis 4	No abnormalities	Absent	No
Acontias occidentalis 5	No abnormalities	Absent	No
Aparallactus capensis 1	Slight lung decay	Absent	No
Aparallactus capensis 2	No abnormalities	Present	No
Chamaeleo dilepis	No abnormalities	Present	No
Gerrhosaurus flavigularus	Slight lung decay	Absent	Present
Lycophidion capense 1	No abnormalities	Present	Present
Lycophidion capense 2	No abnormalities	Absent	No
Mochlus sundevallii	No abnormalities	Present	Present
Nucras intertexta	No abnormalities	Present	Present
Psammophis brevirostris	No abnormalities	Absent	No

A. occidentalis, an *A. capensis*, and the *G. flavigularis*. The *G. flavigularus, L. capense, M. sundevalli,* and *N. intertexta* specimens exhibited pulmonary oedema of varying degrees (Fig. 1C). Terminally aspirated carbon particles were present in the respiratory system (trapped in mucosal mucus) of an *A. occidentalis* specimen indicating smoke inhalation. No acute degeneration of brain matter could be identified from cranial cross sections although the brain of a single *L. capense* specimen exhibited engorged blood vessels (Fig. 1D). The lack of histologically-visible injuries to brain tissue, suggests that nervous tissue injury may not be playing a significant role in the cause of death, although temporary unconsciousness caused by fire-induced oxygen deprivation and/ or toxic gas exposure may lead to fatal heat induced nervous system injury as a result of immobilisation. Histopathological evidence suggests terminal heart failure (possibly heat induced cardiac arrest) or toxic gas (HCN and CO) induced pulmonary alveolar injury as a potential cause of death for some of the collected specimens, similar to what has been described for humans in fire induced mortalities (Busuttil, 2008; Gill and Martin, 2015). The presence of deceased soil living reptiles on the surface as they vacated substrate may indicate a possible mechanism transporting heated gasses into the subsurface.

These observations document diagnostic features observed in some reptiles which succumbed to fire conditions during a single management fire. General trends of species and trait-specific susceptibility to fire-induced faunal mortalities across multiple biomes and management regimes can only be established through continuous documentation. Pre-fire surveys quantifying reptile abundance and density may allow for better direct comparisons of mortality rate and the direct impact of fires on reptile population survivorship whilst the continued investigation of cause of death may better illustrate functional group susceptibility to fire effects.

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