

Cytological and Morphological Characteristics of the Spermatozoon of the Desert Lizard *Scincus scincus* (Squamata, Sauropsida)

Características Citológicas y Morfológicas del Espermatozoide del Lagarto del Desierto *Scincus scincus* (Squamata, Sauropsida)

Othman A. Aldokhi¹; Mukhtar Ahmed¹; Eman S. Alenezzy¹ & Abdel Halim Harrath¹

ALDOKHI, O. A.; AHMED, M.; ALENEZY, E. S.; HARRATH, A. H. Cytological and morphological characteristics of the spermatozoon of the desert lizard *Scincus scincus* (Squamata, Sauropsida). *Int. J. Morphol.*, 43(5):1801-1807, 2025.

SUMMARY: In the present study, we aimed to investigate the ultrastructural characteristics of the spermatozoon of the desert lizard *Scincus scincus*. The spermatozoon of this species has a large subacrosomal nuclear space between the acrosome and nucleus. In addition, two ridges are present on either side of the elongated nucleus, which is protected and covered by a nuclear membrane. The nuclear fossa and basal plate are located on the posterior side of the nucleus. Two distinct centrioles are present below the basal plate: proximal and distal centrioles, which are interconnected and provide support to the midpieces and the entire tail region. In longitudinal sections, a microtubular axoneme runs through the tail region and is surrounded by a fibrous sheath and the cytoplasm. In transverse sections, the main piece of the tail appears well-developed and consists of a typical 9+2 axoneme, nine doublets and two central singlets, surrounded by a thick fibrous sheath and plasma membrane. The fibrous sheath is thick at the beginning of the midpiece and gradually thins before completely disappearing at the start of the endpiece. The endpiece is composed of an axoneme surrounded by the plasma membrane.

KEY WORDS: Lizard; Spermatozoon; Acrosome; Flagellum; Cytology; *Scincus scincus*.

INTRODUCTION

Research on reproduction in animal species, especially endangered species that are threatened with extinction, is important for their conservation and restoration (Herrick, 2019; Meretsky *et al.*, 2000). Understanding how these species and those qualified as slow-growing could help preserve the biodiversity (Manlik, 2019). For biodiversity preservation, it is important to improve our knowledge of the reproductive cycle, including mating seasons and gestation periods, of endangered species (Andrabi & Maxwell, 2007).

Climate change has altered habitats and environmental conditions; therefore, it is important to understand how these alterations affect reproduction in animals (Bronson, 2009). Climate change has globally influenced the timing of ecological events, including reproduction and migration patterns, in numerous species (Bronson, 2009). In the present study, we aimed to examine the ultrastructural characteristics of the spermatozoon in a slow-growing species, the desert lizard *Scincus scincus* (Aldokhi *et al.*, 2019).

The process of complete spermatid formation is divided into six stages: formation of the acrosomal complex, nuclear positioning, rearrangement of the neck portion, formation of the centrioles, differentiation of the midpiece, and elongation of the tail region. The morphological and ultrastructural events associated with spermiogenesis have been well described and conserved among some reptiles (Camps & Bargallo, 1977; Al-Dokhi, 2012; Al-Shammari, 2012; Al-Dokhi *et al.*, 2013; Al-Dokhi *et al.*, 2015; Kurohmaru *et al.*, 2021). The primary goal of spermatozoon differentiation is to rearrange various components of the spermatozoon, and this ultimately leads to extensive restructuring and remodeling of the spermatid to produce a complete spermatozoon.

The purpose of this study was to examine spermatid formation and differentiation in *S. scincus*. Furthermore, we explored the morphological events, in order to provide a better understanding of sperm morphogenesis, that is, the cellular processes and structures required for the successful formation of a spermatid, in *S. scincus*.

¹ Department of Zoology, College of Science, King Saud University, P.O. Box 2455, Riyadh 11451, Saudi Arabia.
FUNDED. Ongoing Research Funding Program (ORF-2025-17), King Saud University, Riyadh, Saudi Arabia.

MATERIAL AND METHOD

Animal Collection and Maintenance. Ten male lizards (*S. scincus*) were captured during the period of sexual activity (April and May) (Al-Quraishy 2011), from north-east Riyadh (60 km), Saudi Arabia (25°30'N, 49°40'E). The lizards were housed in separate cages and maintained for short periods in Plexiglass boxes filled with 10 cm of clean sand. To keep the temperature optimum ($23^{\circ}\text{C} \pm 1.5^{\circ}\text{C}$), the sand was sprinkled with water. The lizards had ad libitum access to meal worms and water (Al-Quraishy 2011). The lizards were sacrificed under anesthesia according to the ethical guidelines outlined by King Saud University, Saudi Arabia.

Tissue Preparation and Ultrastructural Study. The lizards were euthanized under ether anesthesia and dissected to remove the testis; the tissues were sliced into small cubes (1 mm^3) and fixed in 3 % buffered (0.1 M sodium cacodylate buffer; pH 7.2) glutaraldehyde for 4 h at 4°C . The specimens were then fixed in 1 % osmium tetroxide (OsO_4) for 1 h 30 min. Dehydration of the tissues was performed using an ascending gradient of ethanol and cleared in propylene oxide before embedding in pure resin (SPI, Toronto, Canada) (Reynolds 1963). The tissues were cut into ultrathin sections (70–85 nm) using an ultramicrotome (UCT; Leica, Germany) with a diamond knife (Diatome, Biel, Switzerland), placed on 300 mesh copper grids, and stained with uranyl acetate (20 min) and lead citrate (5 min). The sections were photographed using a transmission electron microscope (JEOL JEM-1011) operated at 80 kV using Tengra™ software at the Central Laboratory, King Saud University, Riyadh, Saudi Arabia. The electron micrographs were digitized and finalized using the Adobe Photoshop software.

RESULTS

Spermatid differentiation in *S. scincus* can be divided into six stages: formation of the acrosomal complex, nuclear positioning, rearrangement of the neck portion, formation of the centrioles, differentiation of the midpiece, and elongation of the tail region (Fig. 1A). In this study, the spermatozoon head was found to be elongated, slender, and slightly curved at the center of the nucleus (Fig. 1B). The acrosome complex consists of the main acrosome, which is covered with an acrosomal cap, and subacrosomal nuclear space. The nuclear region consists of an elongated and flattened nucleus surrounded by the plasma membrane. The entire sperm head is surrounded by a layer of cytoplasm. At the base of the head region is a distinct convex curve enclosing the implantation fossa (Fig. 1C). In the mid-tail region, there are two distinct centrioles connected to a long axoneme surrounded by a layer of thick fibrous sheath and thin layer of cytoplasm (Fig. 1D).

Acrosomal Complex. The acrosomal complex is located in the apical region of the sperm head and consists of a conical, elongated vesicular membrane. The acrosome is lean and envelopes the tapered anterior end of the nucleus (Figs. 2A and 2B). The base of the acrosomal complex rests on the widening region of the nucleus with a distinct shoulder-like appearance. The subacrosomal cone lies deep within the acrosomal cortex. It lacks a membrane and forms a thick layer. The acrosome is circular at its base and becomes cone-shaped in the sagittal section near the apical/anterior tip. The space between the nuclear envelope and vesicular membrane (subacrosomal nuclear space) contains dense materials (Fig. 1C). Within the acrosomal complex, there are no distinct acrosomal membranes, and we observed delineation only between the electron-dense and lucent regions (Fig. 1B, arrows). Two distinct acrosomal lucent ridges were observed on either side of the nucleus (Fig. 2B, arrows). A perforatorial base plate is also present, as observed in other lizards. These components together form an acrosomal complex (2C).

Nucleus. In the longitudinal section, the nucleus is slender, elongated, and slightly curved at the center. It tapers anteriorly within the acrosomal complex and appears conical (Figs. 1A and 1B). The nucleus contains highly condensed chromatin material and is surrounded by a nuclear membrane. The nucleus is flat on the posterior side and has a compact convex shape on the implantation side. The implantation fossa is connected to the proximal centriole (Fig. 1C). In cross-section, the nucleus is circular and surrounded by a cylindrical layer of manchette microtubules (Fig. 2E).

Neck Region and Centrioles. The neck region is distinctly demarcated by a boundary at the base plate between the sperm head and tail, forming a junction between the nucleus and midpiece (Fig. 1C). Furthermore, the junction houses centrioles (Fig. 1C) consisting of nine triplets with two central singlets (Fig. 3B-D). This junction consists of two centrioles and deposits of pericentriolar materials. The proximal centriole is positioned centrally parallel to the base of the nucleus. The distal centriole constitutes the basal body of the axoneme. The long axis of the distal centriole, which forms the basal body of the flagellum, lies along the long axis of the axoneme. The distal centriole is perpendicular to the proximal centriole. An extensive deposit of perinuclear material extends from the nuclear fossa to cover the proximal centriole (Fig. 1C). During this process, the two centrioles steadily move to the opposite pole of the nucleus (Fig. 1C). As differentiation proceeds, a nuclear invagination appears on the posterior surface of the nucleus; with further differentiation, the invagination deepens and forms an arch-shaped structure, known as the implantation fossa (Fig. 1C).

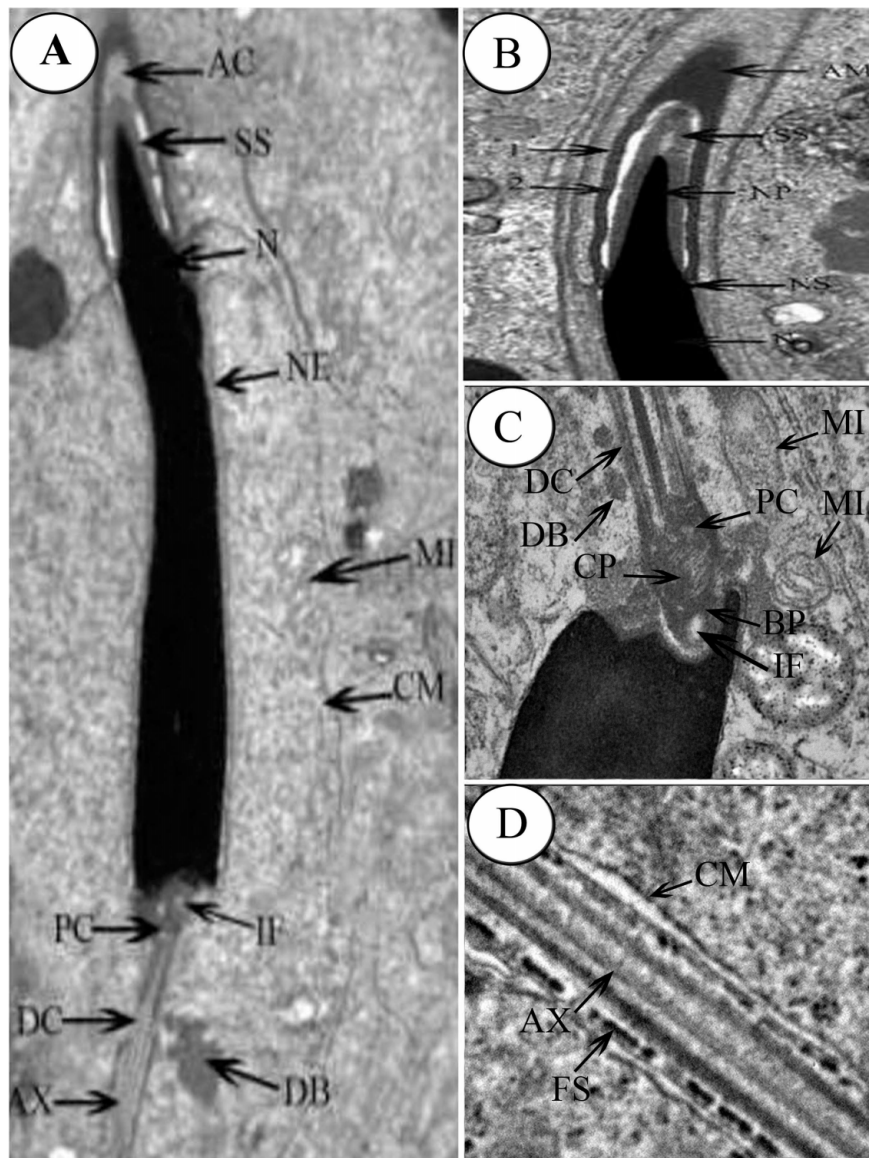


Fig. 1. A. An elongated spermatozoon showing features such as the head, midpiece, and anterior part of the tail region. The head has a well-developed acrosome (AC) covered with an acrosomal cap. There is a distinct subacrosomal space (SS) between the acrosome and anterior nucleus (N). The N is covered with a nuclear envelope (NE). The entire spermatozoon is surrounded by a few mitochondria (MI). At the end of the N, there is a groove (implantation/nuclear fossa) (IF). Beneath the implantation fossa are the proximal (PC) and distal centrioles (DC). A small portion of the axoneme (AX) can also be observed in the later phase. A few scattered dense bodies (DB) are present in the tail region (5000 $\times$). B. An enlarged portion of the anterior head showing all membranes. There are two distinct acrosomal membranes, labelled as 1 and 2. At the tip of the head is the AC. There is a distinct SS. A cone-shaped nuclear apex (NP), which is dark in color, is shown. Two distinct acrosomal lucent ridges are present in the N; they are opposite to each other (NS) and terminate at the apex of the nucleus (8000 $\times$). C. In the midpiece of the spermatozoon, the implantation fossa (IF) is positioned at the basal plate (BP). Beneath the basal plate is a connecting piece (CP), which connects the proximal and distal centrioles (PC and DC). A large number of MI and few DB can also be noted (8000 $\times$). D. A magnified longitudinal section of the tail region showing fibrous sheath (FS), which is surrounded by the cytoplasm (CM). The AX is present at the center (20000).

The implantation fossa is lined by an electron-dense layer—the basal plate, which connects the proximal centriole to the nucleus. The proximal centriole fits in the shallow invagination on the posterior surface of the nucleus (the implantation fossa), whereas the distal centriole is angled at 80° with the proximal centriole parallel to the long axis of the cell. The nuclear envelope surrounds the centriolar complex in a crescent (arch) shape, which may facilitate strong connections (Fig. 1C).

Midpiece. The midpiece consists of the neck region and axoneme surrounded by numerous mitochondria (Fig. 1C). It is much smaller than the head and terminates posteriorly in a distinct annulus. The mitochondria in the cytoplasm gradually begin to accumulate around the axoneme of the midpiece. The mitochondria are uniform and arranged

around the axoneme in the midpiece. Later, the first crown connects to the basal plate and the last crown is attached to the annulus (Fig. 1C).

The distal centriole gives rise to the axoneme and the flagellum is formed by the axoneme, extending throughout the length of the spermatozoon. In successive phases, the distal centriole develops as an electron-dense material that is close to the plasmalemma, where the flagellum emerges from the cell (Fig. 1C). It is organized in the usual 9+2 microtubule pattern surrounded by peripheral fibers (Fig. 1D).

At the distal end of the midpiece, a small dense ring, which appears circular to oval in the cross-section, defines the terminus of the midpiece. It is close to the inner surface of the plasma membrane (Fig. 3B-D).

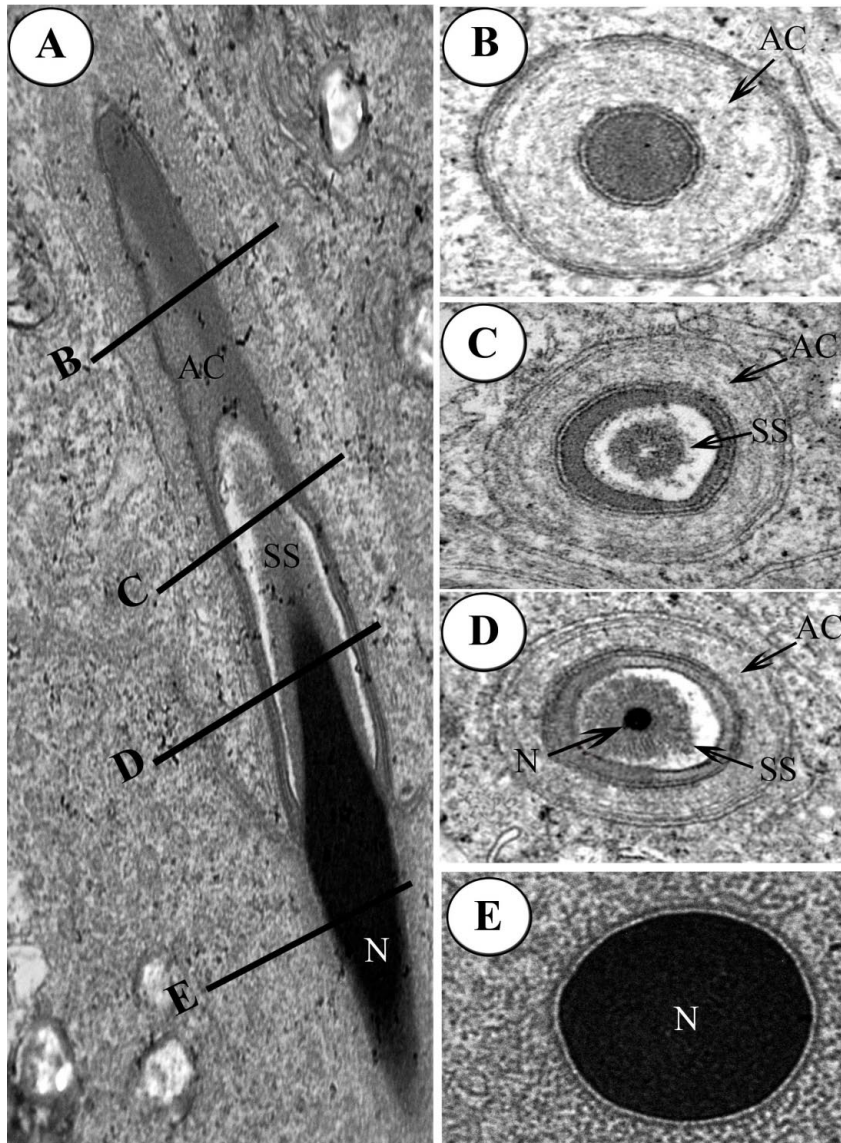


Fig. 2. A. A high-resolution longitudinal section of the head showing four different zones (1–4). Zone (1) represents the acrosomal part, zone (2) represents the subacrosomal space (SS), zone (3) represents the anterior part of the nucleus (N), and zone (4) represents the N (20000×). B. A cross section showing the acrosome (AC) and inner and outer acrosomal membranes (1 and 2) (40000×). C. A cross section showing the SS, AC, and inner and outer acrosomal membranes (1 and 2) (40000×). D. A cross section showing the N, SS, and AC (40000×). E. A cross section showing the dark N with chromatin materials and the nuclear envelope (40000×).

Principal Piece. The initial portion of the principal piece is identified by a reduction in the diameter of the flagellum. It begins immediately after the annulus. This is the longest portion of the flagellum, consisting of the axoneme surrounded by the fibrous sheath, cytoplasm, and plasma membrane (Fig. 1D; Fig. 3B-D). In this region, the axoneme presents the 9+2 microtubule pattern. The diameter of the principal piece gradually reduces, because of a decrease in the cytoplasm content of the fibrous sheath, and the plasma membrane shrinks with a reduction in the width of the fibrous sheath (Fig. 1A; Fig. 3B-D).

Endpiece. The axoneme extends behind the fibrous sheath as an endpiece. The endpiece is referred to as the very slender tail of the spermatozoon. It consists of an axoneme and a plasma membrane (Fig. 3D). The fibrous sheath is noticeably

absent from the endpiece. The pattern of microtubules is largely maintained, although their diameters are significantly reduced.

DISCUSSION

Morphological evolution of spermatids in most lizards have resulted in significant differences in size, location, and partition (Talbot, 1991). The manifestation of acrosomal layers enclosing the nuclear tip is common in all squamates (Da Cruz-Landim & Da Cruz-Höfling, 1977): a uniform electron-dense acrosome is present, which is similar to that in *S. scincus*. The sperm head has a blunt-pointed acrosome, a subacrosomal cone, and a conical nuclear tip of a flattened and elongated nucleus, which lies mostly within this cone membrane. The initial development of the acrosome

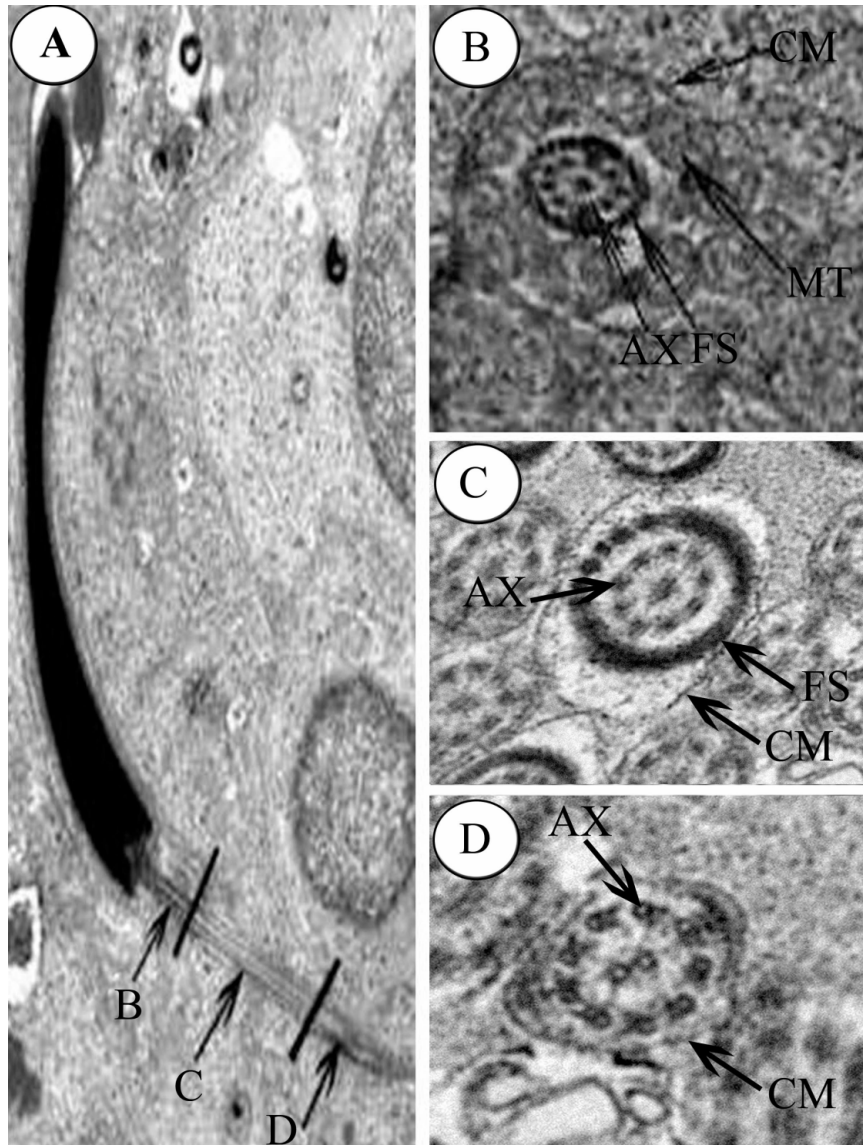


Fig. 3. A. A longitudinal section showing the entire spermatozoon. The tail has three different regions, viz., mid, principle, and end parts (1–3) (10000×). B. A cross section of the anterior (mid) tail showing the axoneme (AX) and fibrous sheath (FS) surrounded by the cytoplasm (CM). A ring of round mitochondria (MI) can be observed between the FS and CM (40000×). C. A cross section of the principle tail region showing the AX, FS, and CM (40000×). D. A cross section of the endpiece showing the AX and CM, which clearly lacks the FS (60000×).

complex in *S. scincus* testes is similar to that in many other squamates (limbed) and other reptiles (Dehlawi *et al.*, 1993; Dehlawi & Ismail, 1994; AlDokhi, 1996; Al-Dokhi, 2012; Al-Dokhi *et al.*, 2013). The dark-flattened nucleus of *S. scincus* has evenly distributed chromatin, with minor heterochromatin clustering, which is different from the heterochromatic nuclei of other species, such as *Sphenodon* (Healy & Jamieson, 1994) and chelonians (Zhang *et al.*, 2007). When acrosomal development and growth are complete, the nuclei of the early elongating spermatids associate with the cell membrane. This contact with the cell membrane flattens the acrosomal vesicle on the surface of the anterior nucleus, which assists in the lateral migration of the acrosomal shoulders over the apical nuclear head. Furthermore, in some reptiles, such as the chelonian *Diplometopon zarudnyi* (Zhang *et al.*, 2007; Al-Dokhi *et al.*, 2013; Al-Dokhi *et al.*, 2015) and in some crocodilians (Wang *et al.*, 2008), the endonuclear canal extends deep into the nuclear body in middle-elongating spermatids. *Diplometopon zarudnyi* has a visible perforatorium within the endonuclear canal (Al-Dokhi *et al.*, 2013), similar to that observed in other lizards (Saita *et al.* 1987). In contrast, all squamates, including *S. scincus* have an extranuclear perforatorium located in the subacrosomal space within the spermatozoa (Gribbins *et al.*, 2007; Gribbins *et al.*, 2010; Rheubert *et al.*, 2010).

The nucleus, exhibiting the condensation of chromatin, favors maturity and mobility and shields the genome from any external and internal modifications during transport and storage (Krause, 1996). During spermiogenesis, this thick, flattened, and elongated nucleus

is well-recognized by its manchette (Soley, 1994) and by DNA and protein aggregation (Fawcett *et al.*, 1971). The spermatozoa of all lizards including *S. scincus* are slender, except for that of *Eugongylus* (Jamieson & Scheltinga, 1993) which possess spermatozoa of a larger diameter. The centrioles assist in axoneme formation, and this process is comparable in all squamates, including *S. scincus* (Al-Hajj *et al.*, 1987; Phillips & Asa, 1993; Al-Dokhi *et al.*, 2015). Nine peripheral dense fibers run straight towards the axoneme (nine doublets). Another distinctive feature of Squamata is the presence of a small distal centriole; however, it does not extend along the entire midpiece, rather ends well above the annulus within the layer of encircling mitochondria, as in *D. zarudnyi* (Al-Dokhi *et al.*, 2015). In contrast, some reptiles have long distal centrioles (Healy & Jamieson, 1992; Jamieson & Healy, 1992; Oliver *et al.*, 1996).

In Squamata, the spermatozoa present linear mitochondrial cristae, inter mitochondrial dense bodies, a short distal centriole, and a fibrous sheath beginning from the midpiece. Enlarged peripheral fibers of the distal centriole and axoneme are characteristic of all reptiles (Jamieson *et al.*, 1997; Al-Dokhi *et al.*, 2015). The fibrous sheath has malleable properties that can improve spermatozoon mobility (Fawcett, 1970). The annulus, which is covered by a fibrous sheath, is found in *S. scincus*, similar to that in other lizards (Baccetti & Afzelius, 1976; Al-Dokhi *et al.*, 2015). The annulus generally consists of filamentous subunits that bind to the plasma membrane. Its main purpose is to withhold mitochondrial disarticulation from the midpiece during flagellar movement (Fawcett, 1970; Al-Dokhi *et al.*, 2015).

The implantation fossa is well settled on the basal plate. Subsequently, in the same axis, the proximal centriole fits beneath the connecting piece. The distal centriole is involved in the formation of flagellar microtubular components. Other reptiles show different binding abilities; for instance, *Mauremys caspica* (Al-Dokhi & Al-Wasel, 2001; Al-Dokhi & Al-Wasel, 2002) develops an arch-shaped implantation fossa to ensure strong binding. Similar to that of other reptiles and limbed lizards, the midpiece of *S. scincus* sperm tail contains a mitochondrial sheath and an axonemal core, and is completed by an annulus. Mitochondrial pairing in the midpiece of sperm tail is a common feature of reptiles and all vertebrates. In the present study, the mitochondria in the midpiece paired via inter-mitochondrial dense bodies. Similar dense inter-mitochondrial bodies have been observed in other lizards, such as *B. tuberculatus* (Al-Dokhi, 2004) and *A. boskinus* (Al-Dokhi, 2012). The inter-mitochondrial dense bodies are a characteristic of Squamata (Jamieson, 1995).

In conclusion, we described morphological reorganization (using electron microscopy) of spermatids in *S. scincus*. The initial structures of the acrosomal complex are similar, except for sharp acrosomal lucent ridges on either side of the nucleus. Nucleus formation is similar to that observed in other reptiles. In the tail region, the observed changes were similar to those observed in all other squamates. The findings of this study can be used to inter- and intra-link the phylogenetic and evolutionary aspects of other reptiles.

ALDOKHI, O. A.; AHMED, M.; ALENEZY, E. S.; HARRATH, A. H. Características citológicas y morfológicas del espermatozoide del lagarto del desierto *Scincus scincus* (Squamata, Sauropsida). *Int. J. Morphol.*, 43(5):1801-1807, 2025.

RESUMEN: En el presente estudio, nuestro objetivo fue investigar las características ultraestructurales del espermatozoide del lagarto del desierto *Scincus scincus*. El espermatozoide de esta especie presenta un amplio espacio nuclear subacrosómico entre el acrosoma y el núcleo. Además, se observan dos crestas a ambos lados del núcleo alargado, el cual está protegido y cubierto por una membrana nuclear. La fosa nuclear y la placa basal se ubican en la parte posterior del núcleo. Por debajo de la placa basal se encuentran dos centriolos distintos: centriolo proximal y distal, que están interconectados y proporcionan soporte a la pieza intermedia y a toda la región caudal. En cortes longitudinales, un axonema microtubular atraviesa la región de la cola, rodeado por una vaina fibrosa y el citoplasma. En cortes transversales, la pieza principal de la cola aparece bien desarrollada y consiste en un axonema típico 9+2, con nueve dobletes y dos singletes centrales, rodeados por una gruesa vaina fibrosa y la membrana plasmática. La vaina fibrosa es gruesa al inicio de la pieza intermedia y se adelgaza progresivamente hasta desaparecer por completo al comienzo de la pieza terminal. La pieza terminal está compuesta por un axonema rodeado por la membrana plasmática.

PALABRAS CLAVE: Lagarto; Espermatozoide; Acrosoma; Flagelo; Citología; *Scincus scincus*.

REFERENCES

- Aldokhi, O. A.; Alwasel, S. & Harrath, A. H. Ultrastructural and histochemical study of previtellogenic oogenesis in the desert lizard *Scincus mitranus* (Squamata, Sauropsida). *J. Morphol.*, 280(3):381-94, 2019.
- Al-Dokhi, O. An electron microscope study of sperm tail differentiation of the lizard, *Acanthodactylus boskinus* (Squamata: Reptilia). *Int. J. Zool. Res.*, 8(3):121-9, 2012.
- Al-Dokhi, O. Electron microscopic study of sperm tail differentiation in the lizard *Bunopus tuberculatus* (Squamata, Reptilia). *Saudi J. Biol. Sci.*, 11:111-7, 2004.
- AlDokhi, O. Ultrastructural studies on the differentiation of the sperm head in the sand skink *Scincus mitranus* (Anderson 1871) (Squamata, Reptilia). *Arab. Gulf J. Sci. Res.*, 14:471-82, 1996.
- Al-Dokhi, O.; Ahmed, M.; Al-Dosary, A. & Al-Sadoon, M. K. Ultrastructural study of spermiogenesis in a rare desert amphibaenian *Diplometopon zarudnyi*. *C. R. Biol.*, 336(10):473-8, 2013.
- Al-Dokhi, O. & Al-Wasel, S. Ultrastructure of spermiogenesis in the freshwater turtle *Mauremys caspica* (Chelonia, reptilia), I-The acrosomal vesicle and the endonuclear canals formation. *J. Egyptian German Soc. Zool.*, 36:93-106, 2001.

- Al-Dokhi, O. & Al-Wasel, S. Ultrastructure of spermiogenesis in the freshwater turtle *Mauremys caspica* (Chelonia, Reptilia). III. Sperm tail formation. *J. Union Arab. Biol. Zool.*, 18: 327-41, 2002.
- Al-Dokhi, O.; Mukhtar, A.; Al-Dosary, A. & Al-Sadoon, M. Ultrastructural differentiation of sperm tail region in *Diplometopon zarudnyi* (an amphibaenian reptile). *Saudi J. Biol. Sci.*, 22(4):448-52, 2015.
- Al-Hajj, H.; Janakat, S. & Mahmoud, F. Electron microscopic study of sperm head differentiation in the lizard *Agama stellio*. *Can. J. Zool.*, 65:2959-68, 1987.
- Al-Quraishy, S. A new *Choleoeimeria* species (Apicomplexa: Eimeriidae) infecting the gall bladder of *Scincus mitranus* (Reptilia: Scincidae) in Saudi Arabia. *J. Parasitol.*, 97(6):1125-8, 2011.
- Al-Shammari, A. M. Additional records of lizards in Ha'il Province, Saudi Arabia. *Russ J. Herpetol.*, 19(4):287-91, 2012.
- Andrabi, S. & Maxwell, W. A review on reproductive biotechnologies for conservation of endangered mammalian species. *Anim. Reprod. Sci.*, 99(3-4):223-43, 2007.
- Baccetti, B. & Afzelius, B. A. The biology of the sperm cell. *Monogr. Dev. Biol.*, (10):1-254, 1976.
- Bronson, F. H. Climate change and seasonal reproduction in mammals. *Philos. Trans. R. Soc. Lond. B. Biol. Sci.*, 364(1534):3331-40, 2009.
- Camps, J. and Bargallo, R., 1977. Espermatozoos de reptiles. I. Ultraestructura de los espermatozoides de *Elaphe scalaris* (Schinz). *Bol R Soc Esp Hist Nat Secc Biol*, 75: 439-46.
- Da Cruz-Landim, C. & Da Cruz-Höfling, M. A. Electron Microscope Study of Lizard Spermiogenesis in *Tropidurus Torquatus* (Lacertilia). *Caryologia*, 30(2):151-62, 1977.
- Dehlawi, G. & Ismail, F. Studies of the ultrastructure on the spermiogenesis of Saudian reptiles. 13-The sperm differentiation in the snake. *Proceedings Zoological Soc. A. R. Egypt*, 25:5-106, 1994.
- Dehlawi, G., Ismail, M. & Sarhan, O. Ultrastructure of the spermiogenesis of saudian reptiles 8 sperm head differentiation in *Agama blandfordi*. *Mol. Androl*, 5:105-14, 1993.
- Fawcett, D. W. A comparative view of sperm ultrastructure. *Biol. Reprod. Suppl.*, 2:90-127, 1970.
- Fawcett, D. W.; Anderson, W. A. & Phillips, D. M. Morphogenetic factors influencing the shape of the sperm head. *Dev. Biol.*, 26(2):220-51, 1971.
- Gribbins, K. M.; Mills, E. M. & Sever, D. M. Ultrastructural examination of spermiogenesis within the testis of the ground skink, *Scincella laterale* (Squamata, Sauria, Scincidae). *J. Morphol.*, 268(2):181-92, 2007.
- Gribbins, K. M.; Rheubert, J. L.; Anzalone, M. L.; Siegel, D. S. & Sever, D. M. Ultrastructure of spermiogenesis in the cottonmouth, *Agkistrodon piscivorus* (Squamata: Viperidae: Crotalinae). *J. Morphol.*, 271(3):293-304, 2010.
- Healy, J. & Jamieson, B. Ultrastructure of the spermatozoon of the tuatara (*Sphenodon punctatus*) and its relevance to the relationships of the Sphenodontida. *Philos. Trans. R. Soc. Lond. B Biol. Sci.*, 335(1274):193-205, 1992.
- Healy, J. & Jamieson, B. The ultrastructure of spermatogenesis and epididymal spermatozoa of the tuatara *Sphenodon punctatus* (Sphenodontida, Amniota). *Philos. Trans. R. Soc. Lond. B Biol. Sci.*, 344(1308):187-99, 1994.
- Herrick, J. R. Assisted reproductive technologies for endangered species conservation: developing sophisticated protocols with limited access to animals with unique reproductive mechanisms. *Biol. Reprod.*, 100(5):1158-70, 2019.
- Jamieson, B. G. M. *The ultrastructure of spermatozoa of the Squamata (Reptilia) with phylogenetic considerations*. In: Jamieson, B. G. M.; Ausio, J. & Justin, J. L. (eds). *Advances in Spermatozoal Phylogeny and Taxonomy*. Mém natn Hist nat Paris, Paris, 166:359-83, 1995.
- Jamieson, B. G. M. & Healy, J. The phylogenetic position of the *Tuatara sphenodon* (Sphenodontida, Amniota), as indicated by cladistic analysis of the ultrastructure of spermatozoa. *Philos. Trans. R. Soc. Lond. B Biol. Sci.*, 335(1274):207-19, 1992.
- Jamieson, B. G. M. & Scheltinga, D. M. The ultrastructure of spermatozoa of *Nangura spinosa* (Scincidae, Reptilia). *Mem. Queensl. Mus.*, 34(1):169-79, 1993.
- Jamieson, B. G. M.; Scheltinga, D. & Tucker, A. The ultrastructure of spermatozoa of the Australian freshwater crocodile, *Crocodylus johnstoni* Krefft, 1873 (Crocodylidae, Reptilia). *J. Submicrosc. Cytol. Pathol.*, 29(2):265-74, 1997.
- Krause, W. J. *Meiosis and male reproductive organs. Essentials of human histology*. New York, Little, Brown and Company, 1996. pp 331-52.
- Kurohmaru, M.; Matsui, T.; Igarashi, H.; Hattori, S. & Hayashi, Y. Postnatal testicular development and actin appearance in the seminiferous epithelium of the Habu, *Trimeresurus flavoviridis*. *Anat. Histol. Embryol.*, 50(2):417-21, 2021.
- Manlik O. The importance of reproduction for the conservation of slow-growing animal populations. *Adv. Exp. Med. Biol.*, 1200:13-39, 2019.
- Meretsky, V. J.; Snyder, N. F.; Beissinger, S. R.; Clendenen, D. A. & Wiley, J. W. Demography of the *California condor*: implications for reestablishment. *Conserv. Biol.*, 14(4):957-67, 2000.
- Oliver, S. C.; Jamieson, B. G. M. & Scheltinga, D. M., The ultrastructure of spermatozoa of Squamata. II. Agamidae, Varanidae, Colubridae, Elapidae, and Boidae (Reptilia). *Herpetol.*, 52(2):216-41, 1996.
- Phillips, D. M. & Asa, C. S. *Strategies for formation of the midpiece*. In: Baccetti, B. (ed.) *Comparative spermatology 20 years after*. New York, Raven Press., 1993. pp 997-1000.
- Reynolds, E. S. The use of lead citrate at high pH as an electron-opaque stain in electron microscopy. *J. Cell Biol.*, 17(1):208-12, 1963.
- Rheubert, J.; Wolf, K. W.; Wilson, B. & Gribbins, K. Ultrastructure of spermiogenesis in the Jamaican Anole, *Anolis lineatopus*. *Acta Zool.*, 91:373-496, 2010.
- Saita, A.; Comazzi, M. & Perrotta, E. *Electron microscope study of spermiogenesis in Caiman crocodylus L.* *Bollettino Di Zoologia*, 54(4):307-18, 1987.
- Soley, J. Centriole development and formation of the flagellum during spermiogenesis in the ostrich (*Struthio camelus*). *J. Anat.*, 185(Pt 2)(Pt 2):301-13, 1994.
- Talbot, P. *Compartmentalization in the acrosome. Comparative Spermatology 20 Years After*. Raven Press, New York, 1991, pp 225-59.
- Wang, L.; Wu, X.; Xu, D.; Wang, R. & Wang, C. Development of testis and spermatogenesis in *Alligator sinensis*. *J. Appl. Anim. Res.*, 34(1):23-8, 2008.
- Zhang, L.; Han, X. K.; Li, M. Y.; Bao, H. J. & Chen, Q. S. Spermiogenesis in soft-shelled turtle, *Pelodiscus sinensis*. *Anat. Rec. (Hoboken)*, 290(10):1213-22, 2007.

Corresponding author:
Prof. Abdel Halim Harrath
Department of Zoology
College of Science
King Saud University
11451 Riyadh
SAUDI ARABIA

E-mail: hharrath@ksu.edu.sa

ORCID: <http://orcid.org/0000-0002-2170-1303>