

**AN IMMUNOHISTOCHEMICAL AND ULTRASTRUCTURAL
STUDY OF THE OVARY OF THE IMMATURE OSTRICH
(*STRUTHIO CAMELUS*)**

By

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Table of contents

Acknowledgement.....	ii
List of figures.....	v
List of tables.....	xi
Summary	xii
Declaration	xiv
Foreword.....	xv
CHAPTER ONE	
General introduction	1
CHAPTER TWO	
The gross anatomical and histological structure of the ovary in the sexually immature ostrich.....	12
CHAPTER THREE	
Ultrastructural features of healthy and atretic ovarian follicles in the sexually immature ostrich.....	44
CHAPTER FOUR	
An immunohistochemical localization of intermediate filament proteins in the ovary of the sexually immature ostrich.....	78
CHAPTER FIVE	
The distribution of progesterone, oestrogen and androgen receptors in the ovary of the sexually immature ostrich.....	96
CHAPTER SIX	
Immunoreactivities to protein gene product 9.5, neurofilament protein and neuron specific enolase in the ovary of the sexually immature ostrich.....	114
Appendix I.....	140
Appendix II.....	141

List of figures

Figure 1.1: Climatic conditions in South Africa.....	7
Figure 2.1: Active ovary of an immature ostrich.....	34
Figure 2.2: Active ovary showing healthy and atretic follicles.....	34
Figure 2.3: Inactive ovary showing a granular surface.....	35
Figure 2.4: Portion of the ovary showing the cortex and medulla.....	35
Figure 2.5: Primordial follicle.....	36
Figure 2.6: Early previtellogenic follicle.....	36
Figure 2.7: Early previtellogenic follicle showing yolk nucleus.....	37
Figure 2.8: Late previtellogenic follicle.....	37
Figure 2.9: Late previtellogenic follicle showing differentiated thecal gland cells in the thecal layer.....	38
Figure 2.10: Vitellogenic follicle.....	38
Figure 2.11: Higher magnification of vitellogenic follicle demonstrating yolk vesicles in the oocyte.....	39
Figure 2.12: Vitellogenic follicle showing well-differentiated theca interna and theca externa.....	39
Figure 2.13: Portion of the cortex demonstrating atretic primordial follicles...	40
Figure 2.14: Atretic previtellogenic follicle.....	40
Figure 2.15: Atretic previtellogenic follicle demonstrating vacuoles in the oocyte.....	41
Figure 2.16: Portion of an atretic previtellogenic follicle showing a multilayered granulosa cell layer.....	41
Figure 2.17: Atretic vitellogenic follicle type I.....	42
Figure 2.18: Atretic vitellogenic follicle type I (Advanced stages).....	42

Figure 2.19: Atretic vitellogenic follicle type II.....	43
Figure 2.20: Portion of the stroma occupied by stromal interstitial glands.....	43
Figure 3.1: Electron micrograph of healthy primordial follicle.....	66
Figure 3.2: Balbiani's vitelline body.....	66
Figure 3.3: A higher magnification electron micrograph of Balbiani's vitelline body.....	66
Figure 3.4: Granulosa cell in primordial follicle.....	66
Figure 3.5: Basal lamina in primordial follicle.....	67
Figure 3.6: Electron micrograph of healthy previtellogenic follicle.....	67
Figure 3.7: Electron micrograph of late healthy previtellogenic follicle.....	67
Figure 3.8: Higher magnification of <i>zona radiata</i>	67
Figure 3.9: Electron micrograph of early healthy previtellogenic follicle.....	68
Figure 3.10: Electron micrograph of late healthy previtellogenic follicle.....	68
Figure 3.11: Granulosa layer of late healthy previtellogenic follicle.....	68
Figure 3.12: Type I granulosa cells.....	69
Figure 3.13: Type II granulosa cells.....	69
Figure 3.14: Granulosa cell showing apical cytoplasmic processes.....	69
Figure 3.15: Granulosa cell exhibiting transosomes on the apical region.....	69
Figure 3.16: Desmosomes between two granulosa cells.....	70
Figure 3.17: Electron micrograph of transosomes.....	70
Figure 3.18: Basal lamina in previtellogenic follicle.....	70
Figure 3.19: A survey electron micrograph of late healthy previtellogenic follicle.....	70
Figure 3.20: Higher magnification electron micrograph of fibroblast in the thecal layer.....	71

Figure 3.21: Thecal layer showing spindle shaped fibroblasts.....	71
Figure 3.22: Cellular junctions between fibroblasts in the thecal layer.....	71
Figure 3.23: Undifferentiated thecal gland cell.....	72
Figure 3.24: Portion of theca externa showing differentiated thecal gland cells.....	72
Figure 3.25: Higher magnification electron micrograph of healthy vitellogenic follicle showing part of the oocyte and granulosa cell.....	73
Figure 3.26: A survey electron micrograph of healthy vitellogenic follicle.....	73
Figure 3.27: Granulosa cells of healthy vitellogenic follicle.....	73
Figure 3.28: Healthy vitellogenic follicle showing a distinct theca interna and theca externa.....	74
Figure 3.29: Portion of healthy vitellogenic follicle showing fibroblasts and undifferentiated thecal gland cells arranged in strata.....	74
Figure 3.30: Nerve fibres in the theca externa.....	74
Figure 3.31: Electron micrograph of atretic primordial follicle.....	75
Figure 3.32: Electron micrograph of atretic previtellogenic follicle.....	76
Figure 3.33: Amorphous layer in atretic previtellogenic follicle.....	76
Figure 3.34: Swollen mitochondria in the granulosa cells of atretic previtellogenic follicle.....	76
Figure 3.35: Connective tissue between thecal fibroblasts in the theca externa of atretic previtellogenic follicle.....	76
Figure 3.36: Interstitial gland cells in an atretic vitellogenic follicle.....	76
Figure 3.37: Swollen mitochondria in the fibroblast of atretic vitellogenic follicle.....	76

Figure 4.1: Healthy previtellogenic follicle showing a moderate immunostaining for desmin in the granulosa cells.....	89
Figure 4.2: Healthy vitellogenic follicle showing a moderate immunoreactivity for desmin in the theca interna.....	89
Figure 4.3: Healthy previtellogenic follicle showing strong immunoreactivity for vimentin in the granulosa cells.....	90
Figure 4.4: Healthy vitellogenic showing immunostaining for smooth muscle actin in the theca externa.....	90
Figure 4.5: Atretic vitellogenic follicle showing positive immunostaining for desmin in fibroblast-like cell.....	91
Figure 4.6: Atretic vitellogenic follicle type II showing positive immunostaining for desmin in fibroblast-like cells.....	91
Figure 4.7: Atretic vitellogenic type I showing desmin immunoreactivity in the stroma and in the blood vessel walls.....	92
Figure 4.8: Atretic previtellogenic follicle showing a moderate immunostaining for vimentin in the granulosa cell layer.....	92
Figure 4.9: Atretic vitellogenic follicle type I showing positive immunostaining for vimentin in fibroblast-like cells.....	93
Figure 4.11: Atretic vitellogenic follicle type I showing smooth muscle actin immunoreactivity in fibroblast-like and smooth muscle cells.....	94
Figure 4.12: Atretic vitellogenic follicle type II showing smooth muscle actin immunoreactivity in fibroblasts infiltrating the interstitial mass.....	94
Figure 5.1: A portion of cortex showing oestrogen receptor immunoreactivity in the germinal epithelium.....	111

Figure 5.2: Previtellogenic follicle showing immunoreactivity for oestrogen receptor in the thecal gland cells.....	111
Figure 5.3: Part of the cortex showing immunostaining for progesterone receptor in the germinal epithelium.....	111
Figure 5.4: Blood vessel wall showing immunoreactivity for progesterone receptor in the tunica media.....	111
Figure 5.5: Part of stroma showing a moderate immunoreactivity for progesterone receptor in the fibroblasts.....	112
Figure 5.6: Ovarian cortex showing strong immunoreactivity for androgen receptor in the germinal epithelium.....	112
Figure 5.7: Part of cortex showing positive immunostaining for androgen receptor in the granulosa cells.....	112
Figure 6.1: Neurofilament protein immunoreactive nerve bundles in the ovarian stalk.....	131
Figure 6.2: Neurofilament protein immunoreactive nerves in the medulla and cortex.....	131
Figure 6.3: Neurofilament protein nerve bundles associated with blood vessels.....	132
Figure 6.4: Healthy previtellogenic follicle showing neurofilament protein immunoreactive nerve fibres in the theca interna and stroma.....	132
Figure 6.5: Part of the cortex showing distribution of neurofilament protein immunoreactive nerve fibres.....	133
Figure 6.6: Vitellogenic follicle showing neurofilament protein immunoreactive nerve fibres.....	133

Figure 6.7: Atretic vitellogenic follicle type I showing neurofilament protein immunoreactive nerve fibres in the theca externa.....	134
Figure 6.8: Atretic vitellogenic follicle type I showing nerve fibre within hyalinized connective tissue.....	134
Figure 6.9: Neuron specific enolase immunoreactive bundle in the medulla.....	135
Figure 6.10: Nerve cell body immunoreactive for neuron specific enolase.....	135
Figure 6.11: Late previtellogenic follicle showing immunoreactivity for neuron specific enolase in differentiated thecal gland cells.....	136
Figure 6.12: Vitellogenic follicle showing immunoreactivity for neuron specific enolase in thecal gland cells.....	136
Figure 6.13: Portion of cortex showing protein gene product 9.5 immunoreactivity.....	137
Figure 6.14: Protein gene product 9.5 immunoreactive nerve bundles in the ovarian stalk and medulla.....	137
Figure 6.15: Protein gene product 9.5 immunoreactive nerve bundle associated with a blood vessel in the medulla.....	138
Figure 6.16: Late previtellogenic follicle showing protein gene product 9.5 immunoreactive nerve fibres in the thecal layer.....	138

List of tables

Table 4.1: Summary of the immunohistochemical localization of vimentin, desmin and smooth muscle actin in healthy ovarian follicles of the immature ostrich.....87

Table 4.2: Summary of the immunohistochemical localization of vimentin, desmin and smooth muscle actin in atretic ovarian follicles of the immature ostrich.....88

Table 6.1: Summary of density and distribution of nerve fibres immunoreactive for neurofilament protein, neuron specific enolase and protein gene product in the active ovary of the sexually immature ostrich.....119

Table 6.2: Summary of density and distribution of nerve fibres immunoreactive for neurofilament protein, neuron specific enolase and protein gene product in the regressive ovary of the sexually immature ostrich.....120

SUMMARY

AN IMMUNOHISTOCHEMICAL AND ULTRASTRUCTURAL STUDY OF THE
OVARY OF THE IMMATURE OSTRICH (*STRUTHIO CAMELUS*)

By

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The aim of this study was to investigate the components of the ovary in the sexually immature ostrich by using immunohistochemistry, light microscopy and electron microscopy. The light and electron microscopic studies carried out, revealed that the oocyte in the sexually immature ostrich is surrounded by seven layers which included the *zona radiata*, *lamina perivitellina*, *stratum granulosum*, *basal lamina*, thecal layers (theca interna and theca externa), connective tissue layer and superficial epithelium (see details in Chapter Two and Three). Several morphological and immunohistochemical changes occurred as the follicles developed and regressed, suggesting that ovarian follicles in the sexually immature ostrich undergo a cycle of growth and degeneration as reported in other avian species.

In the present study, thecal gland cells in the ovary of the sexually immature ostrich were common. In addition, interstitial gland cells were a notable feature in atretic follicles as described in the ovary of the crow, common myna and dove (Guraya and Chalana, 1976). Further investigations on the interstitial gland cells will provide an insight into the process of steroidogenesis in the sexually immature ostrich.

As discussed in Chapter five, various cells in the ovary showed immunoreactivity to oestrogen, progesterone and androgen receptors. These observations indicated that the ovarian tissue in the sexually immature ostrich is a potential target for gonadal hormones. Thus, it can be assumed that steroid hormones regulate ovarian functions in the ostrich.

The use of immunohistochemical procedures proved to be an excellent method to investigate the distribution of nerves in the ovary. The results of this study have shown that the ovary in the sexually immature ostrich is well-innervated. However, further studies are required to differentiate between cholinergic and adrenergic nerve fibres.

Declaration

I hereby declare that the work presented here is my original work. To the best of my knowledge, this work has never been published or submitted for a degree in this University. The University of Pretoria reserves the right of permission for duplication of the whole thesis or in part thereof.

.....
W.H. Kimaro

November, 2005.

Foreword

The main reason for conducting the present study was the lack of information on the morphology of the ovary in the sexually immature ostrich. A total of 26 sexually immature female ostriches aged between 12 and 14 months were used in the present study. Ovarian tissue samples were collected during the active reproductive phase (August to February), the regressive reproductive phase (March to early May) and the inactive reproductive phase (Late May to July). Tissue samples were processed routinely for light and electron microscopic studies. Immunohistochemistry was performed on either frozen or paraffin-embedded sections.

The objectives of Chapter Two and Chapter Three were to investigate the histological and ultrastructural organization of the ovary in the sexually immature ostrich. At the light microscope level, healthy and atretic primordial, previtellogenic and vitellogenic follicles were observed. The healthy follicles were composed of an oocyte surrounded by a granulosa cell layer and a thecal layer.

At the electron microscope level, granulosa cells in healthy follicles displayed apical cytoplasmic processes. Attached to the cytoplasmic processes were transosomes. A basal lamina separated the granulosa cell layer from the underlying thecal layer. The basal lamina closest to the granulosa cell layer was more electron dense than that adjacent to the thecal layer. The thecal layer contained undifferentiated (type I) and differentiated (type II) thecal gland cells, as well as vacuolated thecal cells. The type I thecal gland cells

contained a large oval or elongated nucleus, which exhibited clumps of heterochromatin. The nuclei of type II thecal gland cells were round to oblong in shape with a prominent nucleolus.

Non-bursting atresia was observed in all follicular sizes. The granulosa cells of atretic primordial and previtellogenic follicles contained numerous lipid droplets and electron dense bodies. Very few transosomes were observed in atretic follicles. Two forms of atresia were observed in vitellogenic follicles. Type I atresia resulted in the infiltration of the entire follicle by hyalinized connective tissue. In type II atresia, granulosa and theca interna cells differentiated into interstitial gland cells. These results indicate that the structural organization of the ovary in the sexually immature ostrich is similar to that reported in other avian species. In addition, it is apparent that ovarian follicles in the sexually immature ostrich undergo a cycle of growth and degeneration.

The objective of Chapter Four was to study the distribution of the intermediate filaments, desmin, vimentin and smooth muscle actin, in the ovary of the sexually immature ostrich. Positive immunostaining for desmin was observed in the granulosa cells of healthy primordial and previtellogenic follicles. Vimentin immunoreactivity was demonstrated in the granulosa cells of all follicles except the vitellogenic atretic follicles. Fibroblasts in healthy and atretic (type I) follicles exhibited strong immunostaining for smooth muscle actin. The results of this chapter suggested that the distribution of intermediate filaments changes during follicular development and atresia.

The objective of Chapter Five was to determine the distribution of steroid hormone receptors in the ovary of the sexually immature ostrich. Strong immunostaining for the oestrogen receptor, progesterone receptor and androgen receptor was observed in the nuclei of the germinal epithelium. Granulosa cells were immunopositive for the progesterone and androgen receptors, but not for the oestrogen receptor. However, positive immunoreactivity for the oestrogen receptor was exhibited in thecal gland cells. The distribution of steroid hormone receptors in the present study appears to be similar to that described in the domestic fowl.

The objective of Chapter Six was to describe the intrinsic innervation of the ovary using antibodies against neurofilament protein, protein gene product 9.5, and neuron specific enolase. Strong immunostaining for neurofilament protein, protein gene product 9.5 and neuron specific enolase was observed in nerve bundles, which coursed through the ovarian stalk and extended into the medulla and cortex. Neuron specific enolase immunoreactive nerve cell bodies were observed in the ovarian stalk and medulla. In addition, thecal and interstitial gland cells demonstrated neuron specific enolase immunostaining. Based on the results of this immunohistochemical study, it would appear that the distribution of immunoreactive nerve fibres in the ovary of the sexually immature ostrich resembles that of the domestic fowl.