PREFACE
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Over the past two decades, the zebrafish (*Danio rerio*) has emerged as a powerful and versatile genetic model to study the vertebrate development and organogenesis (Nusslein Volhard and Haffter, 1996; Nusslein Volhard and Dahm, 2002). As a model system, this fish has several advantages such as (i) small size (ii) short generation time (iii) optical transparency of the embryo and (iv) *ex utero* development etc. In addition, the genome of this fish has been sequenced completely and is found to have a strong synteny with that of the human genome that has made this fish a close vertebrate model to study human diseases hence, intensely employed in drug/pharmaceutical discovery research (Barbazuk *et al.*, 2000; Alestrom *et al.*, 2006). Zebrafish is also considered as an excellent model for *in vivo* studies of vertebrate germ line development (Yoon *et al.*, 1997; Raz, 2003; Sakai, 2006; Schlueter *et al.*, 2007). Further, more than 1800 zebrafish mutants have been identified so far, providing a rich resource for the critical analysis of genetic control of development, physiology and disease; consequently, more and more laboratories are now enthused to utilize this unique collection of mutants by adapting this fish as a model for research. Thus, breeding, raising and maintaining adult zebrafish in the laboratory is becoming increasingly demanding. Owing to its growing importance it is the need of the hour to have a thorough knowledge about the reproductive physiology of this fish. A critical understanding of the process of
gametogenesis, factors influencing/governing reproduction and the regulatory mechanisms controlling it will be of great relevance for the successful breeding and maintenance of the stock of zebrafish in the laboratory required for research and experimentation.

Germ line stem cells (GSCs) derived from embryonic primordial germ cells (PGCs) form precursors for a constant supply of differentiating germ cells in the adult gonads (Raz, 2002, 2003; Sakai, 2006; Schlueter et al., 2007; Fan et al., 2008; Nakamura et al., 2011). Steroid hormone implications, gene expression profiles and aromatase enzyme modulations during gonadal differentiation, expression of vasa homologue in PGCs and involvement of various gene products (Chemochine receptor-Cxcr4) and growth factor receptor (IGFr) in the PGC migration are reported for zebrafish (Maac, 1964; Yoon et al., 1997; Braat et al., 1999; Knaut et al., 2000; Fenske and Segner, 2004; Schlueter et al., 2007; Jorgensen et al., 2008). All these studies are restricted to embryonic development. The understanding of regulation of proliferation and differentiation of GSC in adult ovary still remains incomplete owing to the paucity of studies.

Production of fertilizable haploid egg by the adult ovary involves a final step of extrusion of polar bodies which is initiated through the process of germinal vesicle break down (GVBD)/oocyte maturation (Masui and Clarke, 1979; Maestro et al., 1999; Schmitt and Nebreda, 2002; Jamnongjit and Hammes, 2005; Picha and Thomas, 2012). The hormonal interplay, signalling molecules and mechanisms
underlying the process of oocyte maturation are now documented for mammals (Masui and Clarke, 1979; Xia et al., 1994; Sirotkin et al., 2000; Kiapekou et al., 2005; Liu et al., 2010). Corresponding studies on fishes are fragmentary and remain elusive for zebrafish.

In recent years owing to the excessive use of pesticides/insecticides and weedicides, aquatic environment is under constant abuse posing a threat to the life of non-target inhabitants of the system by adversely affecting their immune system, homeostasis and reproduction (McLachlan and Arnold, 1996; Lynn, 2001; Zhou et al., 2009).

The present investigation is an attempt to address few of these questions and to elucidate the processes that determine the fecundity of a female fish such as (i) oogonial proliferation - the rate of oogonial mitosis and their differentiation in the sub adult/adult ovary and the factors (energy sources, metabolic hormones, gonadotropins and growth factors) that influence and possible mechanisms involved in the regulation of these processes using in vitro and in vivo systems (ii) oogenesis - rate of production of oocytes from oogonia (iii) folliculogenesis - development and growth of follicle with special reference to development and differentiation of follicular envelopes (iv) vitellogenesis - source of yolk granules, modes of incorporation of yolk into the oocytes and histochemical analysis of yolk (v) oocyte maturation, hormones, growth factors and endocrine disrupters that influence or alter or inhibit this process using an in vitro system.
The thesis is divided into the following sections:

(i) Introduction
(ii) Materials and methods
(iii) Results
(iv) Discussion
(v) General Summary
(vi) Literature cited

1. Introduction

In this section, an account on the thorough update of literature available, previous investigations undertaken, experimental data prevailing and lacunae existing in our understanding of reproductive processes of the adult zebrafish, are highlighted. The objective of the present investigation is clearly defined. The relevancy of the topics chosen for study, the suitability of the techniques and protocols employed and advantages of use of zebrafish for such an investigation are justified.

2. Materials and methods

This section comprises the details of the materials used and methods employed in the present study. The procurement of the study material, i.e. fish,
hormones (Insulin, Thyroxine, Adrenocorticotropic hormone, Somatotropin, Follicle stimulating hormone, Prolactin, Diethylstilbestrol (DES), growth factors (IGF1), Somatostatin, energy substrate (Galactose), components of culture media (HEPES), chemicals and test chemicals (xenoestrogens) and meteorological (temperature and photophase) data are given. The protocol used for the maintenance of experimental animals and preparation of culture media, oocyte collecting medium, oocyte maturation assay medium, clearing solution, stains, hormone concentrations for in vitro studies, physical parameters used, and duration of the experiments are described. The various endpoints (histology, histochemistry, oocyte maturation assay) and parameters (morphology, morphometry, quantification of oogonia, oocytes, follicular kinetics and rate of germinal vesicle break down (GVBD) recorded are enlisted. The appropriate statistical tools and software packages that have been operationalized in the analysis and presentation of the data are mentioned.

3. Results

In this section, observations made in each of the experiment are sequentially are presented under different subtitles. The text of this section is substantiated with tables, graphs and photomicrographs.
4. Discussion

This section includes a detailed discussion on the results obtained by the present study in comparison with those reported by other workers on similar lines using the same or different model organism. At the end a brief conclusion is included that highlights the relevance and contributions of the present investigation.

5. General Summary

A concise summary of main findings of this study are summarized in this section.

6. Literature cited

The relevant references and reviews used in context with the present study are enlisted in an alphabetical and chronological order.