

# Lesson IV BIOCHEMICAL MODIFICATION OF THE CYTOPLASM

### **1. RNA Accumulation and Function**

Messenger RNA (mRNA), ribosomal RNA (rRNA), and transfer RNA (tRNA)
Polyadenylation of mRNA and its roles (stability, transport, translation)
Utilization of stored RNA in early post-fertilization stages

#### 2. Transition from Oocyte to Mature Gamete

Conclusion of RNA synthesis at the end of the growth phase
Oocyte activation and transition to the zygote
Assembly of the new embryonic genome

### 3. Protein Accumulation and Localization and Meiotic Competence

Role of cell cycle proteins in oocyte development (e.g., Cyclins, CDC25)
Sequestration and translocation of key proteins for meiotic resumption

#### 4. Meiotic Quiescence and Resumption

Inhibitory factors in follicular fluidHormonal stimulation and cell cycle activation

### 5. Growth and Maturation Timelines in Different Mammals

•Differences between rodents, pigs, and larger mammals

# Lesson Va BIOCHEMICAL MODIFICATION OF THE NUCLEUS

## 1. Epigenetic remodelling of chromatin and fuctional endpoints

- Global genome silencing before fertilization
- Gene imprinting and permanent gene expression suppression

# 2. Epigenetic Marks and Chromatin Regulation

•Role of writers, erasers, and readers in modifying chromatin

## **3.** Global DNA Methylation in Oogenesis

•Timeline of DNA methylation during follicular development

•Methods for assessing DNA methylation (immunofluorescence with 5-methylcytosine antibody)

•Correlation between chromatin configuration changes and transcription suppression

## 4. CpG Island Methylation and Genomic Imprinting

For the establishment of germline differentially methylated regions (gDMRs)
 Genomic imprinting: parental-specific gene expression (approx. 100 genes in humans)

•Examples of paternally imprinted genes (maternally expressed) and maternally imprinted (paternally expressed).

# •5. Methods for Detecting DNA Methylation

•Bisulfite mutagenesis: conversion of unmethylated cytosine into uracil

•First and second-generation sequencing (Sanger, NGS) for methylation analysis

•Third-generation sequencing (Nanopore technology): real-time detection of DNA methylation

# UNIT II – Lesson Vb

#### 1. Role of Imprinted Genes in Mammals

Regulate the synthesis of proteins involved in embryonic and fetal growth
Influence parental behavior (lactation, mother-child interaction)

#### 2. Methylation of Imprinted Genes

•Occurs during gametogenesis and remains stable

### 3. ART and Genetic Imprinting

•ART can alter genetic imprinting, increasing the risk of rare diseases in newborns
•Epidemiological studies suggest a higher risk of imprinting disorders in children conceived via IVF and ICSI

### 4. First Experiment on the Function of Imprinted Genes (Surani, 1984)

Creation of uniparental embryos (androgenetic and gynogenetic)
Results: abnormal development, proving that both parental genomes are necessary
Maternal genes are crucial for embryonic development, while paternal genes are essential for extraembryonic tissues

#### 5. Telomerase Activity and Embryonic Development

Telomerase maintains telomere length, ensuring proper cell division and stability
Its activity is tightly modulated also according to its subcellular localization