Chapter IV

MORPHOLOGICAL, HSITOLOGICAL AND HISTOCHEMICAL OBSERVATIONS ON THE OVARIAN TISSUE OF *MUSCA DOMESTICA* (DIPTERA)
Fig. No. 8: Diagram showing morphology of the ovary in *Musca domestica*
(A) MORPHOLOGICAL OBSERVATIONS ON THE OVARIAN TISSUE OF
*MUSCA DOMESTICA*

In the distal half of the abdomen of the female housefly two disc shaped ovaries are present. The ovaries at the end of oogenesis fill the ventral and lateral regions of the abdomen. Each ovary contains 60-75 ovarioles. The ovariole is divisible into three general zones.

1. THE TERMINAL FILAMENT

A dorsal thread like continuation combining with others to attach the ovarioles to the dorsal diaphragm by a suspensory ligament.

2. THE GERMARIUM

Containing the oogonia which became differentiated into oocytes and nurse cells (trophocytes) and

3. THE VITELLARIUM

Comprising follicles or egg chambers surrounded by a single layer of follicular cells (the follicular epithelium).

(B) HISTOLOGICAL OBSERVATIONS ON THE OVARIAN TISSUE OF
*MUSCA DOMESTICA* IN CONTROL AND EXPERIMENTAL GROUPS

(i) IN CONTROL GROUP

The development of ovaries of control group ingested with balanced diet was normal from 2 days to 6 days in which formation of egg follicles and vitellogenesis were normal as observed in *Musca domestica* by a number of workers and on 6th day the oocytes attain such maturity which is required for egg laying (Fig. No. 66 to 69), so, after egg laying yellow colour bodies (corpora lutea) were observed at the base of ovariole showed the normal egg laying while in experimental groups the
development of most of the egg follicles inhibited due to nutritional stress and may be described in the following headings:

CORPUS LUTEUM

After ovulation, the ovary of *Musca domestica* showed the presence of yellow bodies (Corpora lutea) at the junction of the ovariole and the pedicle. In the corpus luteum the tunica propria was strikingly folded. Folding and refolding of the tunica propria was a sign of progressive contraction of the empty follicle, which was soon resorbed when the oocyte has been discharged from the egg follicle. Early signs of degeneration were clearly visible in the follicular epithelium and the remained of the trophocytes, which played an active role in resorption of the corpus luteum. The cellular structure of the follicular epithelium was severely disorganized and marked changes were occurred in the nuclei of the cells. In the early stage the chromatin material was converted to chromatin granules. The nuclei afterwards became pyknotic and were found scattered about irregularly in the cytoplasmic syncytium. In the earlier stage the pyknotic nuclei were many and small but in the later stages their number steadily decreases, while their size increases and they rapidly degenerate. When the next follicle was ready to ovulate the corpus luteum contains a few pyknotic nuclei scattered about in cytoplasmic syncytium. The tunica propria was excessively folded. The cytoplasmic syncytium displayed vacuolation, indicated that active resorption has taken place in the corpus luteum (Fig. No. 107, 108, 109).

(ii) IN EXPERIMENTAL GROUP INGESTED WITH PROTEIN DIET

The ovary of *Musca domestica* has a typical muscid structure. The oogonium was liberated from the germarium at the apex of the polytrophic ovariole and passed into the vitellarium, where it was invested by follicular epithelium. As the follicle and its contents moved down the vitellarium, new follicles were formed successively above it, each presumably contained a single oogonium.

(a) NEWLY EMERGED FLIES

In newly emerged female flies, ingested with protein, the developing egg chambers or follicles were already clearly differentiated. Each oocyte developed in its follicle with the aid of another type of cell (accessory cell). In *Musca domestica* there were two main types of accessory cells e.g. nurse cell or trophocytes and follicle cells. The nurse cells were derived from the oogonium from which the
corresponding oocyte developed. An oogonium undergoes four successive mitotic divisions, resulting in the formation of 16 cells, one of these became an oocyte and the remaining 15 nurse cells. Each follicle was wrapped in follicular epithelium with a tunica propria outside it. The trophocyte cytoplasm, the ooplasm and even the follicular epithelium were seen to contain dense protein positive material.

The nurse cells and oocytes could not be differentiated from each other. Each of them has a nucleus and the trophocytes and oocyte of the same follicle were all interconnected.

(b) TWO DAYS OLD FLIES

The follicle of 2 days old female flies, ingested with protein was much larger and the oocyte was easily distinguishable from the trophocytes. The later have a central nucleus which was larger than the oocyte nucleus and has a thick membrane and coarse chromatin network formed of large, irregularly distributed chromatin granules. Each trophocytes nucleus contains 7-9 prominent nucleoli. The trophocyte nucleoli were rounded. The entire egg follicle was surrounded by multilayered follicular epithelium, whose cells proliferate for some way between the nurse chamber and the developing oocyte. The follicular epithelium rounds the nurse chamber to form only a thin cell layer, because its contents contributed continuously to the growing trophocytes. The nurse cells grown faster than the oocytes and in a short time attain their maximum size. They then receive no further nutrients from the follicular epithelium but continue to supply the developing oocyte from their own stocks of nutrient by way of cytoplasm connections (Fig. No. 70).

(c) FOUR DAYS OLD FLIES

The egg follicle of 4 days old female flies ingested with protein was much larger, as the trophocytes finish their developing. The trophocyte immediately adjacent to the oocyte continued to grow larger than the other trophocytes and it also has a larger nucleus. The trophocyte cytoplasm was highly vacuolated, the chromatin matrial was less dense and there was marked destruction of nuclear material. The nuclear membrane of the nurse cells subsequently broken down and chromatin was released continuously into the cytoplasm, from which nutrients were supplied to the oocyte. There was no partition between the nurse chamber and the oocyte and owing to the absence of follicular epithelium the trophocytes were in
direct contact with the oocyte, so that nutrients can be conveniently transported from
the trophocytes to the oocyte via cytoplasmic bridges.

The ooplasm contains a larger number of irregularly distributed PYP bodies. The
vitellogenin was taken up actively from the haemolymph by micropinocytosis. The
oocyte nucleus was seen to lie quite close to the nurse chamber. The trophocyte
cytoplasm was also vacuolated, with large vacuoles situated near the periphery, i.e.
adjacent to the follicular epithelium. Numerous PYP bodies could be seen scattered
about in the ooplasm (Fig. No. 71).

(d) SIX DAYS OLD FLIES

The rate of oocyte growth in the 6 days old female flies ingested with protein
was high, because the oocyte now attained its full size. The follicular epithelium
was more active, showed that vitellogenin passed actively from the haemolymph to
the ooplasm by micropinocytosis. The nuclei of the follicular epithelial cells were
also more active. The nurse chamber has started to shrink (Fig. No. 72).

The number of trophocytes became smaller in size in comparison to 4 days
old flies. The follicular epithelium was surrounded along the entire egg follicle. It
was composed of a single cell layer. The nuclei of the follicular epithelial cells were
prominent. The fully grown oocyte was filled with PYP bodies which were densely
packed in the centre of the ooplasm and were less dense in the peripheral ooplasm.
Slight vacuolation of the trophocytes was discernible at the periphery and below the
follicular epithelium. The vitelline membrane was also prominent. The chorion was
secreted on the outer surface of vitelline membrane and the inner surface of the
follicular epithelium (Fig. No. 72).

(e) EIGHT DAYS OLD FLIES

The eight days old protein ingested flies showed the presence of large
number of degenerated and disintegrated follicles showed the detachment of chorion
along with follicular epithelium. Due to the formation of large number of resorptive
bodies the flies were unable to laid down the eggs so no corpus luteum has been
observed (Fig. No. 73).

(iii) IN EXPERIMENTAL GROUPS INGESTED WITH CARBOHYDRATE
AND LIPID DIETS

The gradual histological changes in the ovarian tissue of the female flies of
Musca domestica ingested with carbohydrate and lipid diets are as follows:
(a) IN TWO DAYS OLD FLIES INGESTED WITH CARBOHYDRATE AND LIPID DIET

The egg follicles were not much more different in size from that of normal developed follicle. The follicular epithelium surrounded the follicle was single cell layered in thickness with distinct cell boundaries. The cells were mononucleate in nature. The nuclei were large sized in comparison with the cell size. The cells were with small quantity of cytoplasm. The trophocytes were well marked with distinct cell boundaries. They were mononucleate in nature. Some vacuolization has seen in the nucleolasm of the trophocytes.

Trophocytes nucleus was also bigger in size and was not regular in outline. The trophocyte cytoplasm became vacuolated (Fig. No. 74 and 78).

(b) FOUR DAYS OLD FEMALE FLIES OF MUSCA DOMESTICA

The flies fed with carbohydrate and lipid diet showed that the follicular epithelium of the resorbed oocytes was single cell layered in thickness in most of the region of the resorbed oocyte but was double to multilayered at that region which was adjacent to the trophocytes. The cells were with distinct cell boundaries. The nuclei of the follicular epithelium became pycnotic in nature. Some degree of vacuolization has seen in follicular epithelium. The follicular epithelium became multicell layered at the proximal end of the oocyte which was away from the trophocyte region. The degree of vacuolization has increased further in the nucleolasm and the trophocyte cytoplasm. Some vacuoles were very larger in size. The oocyte was mononucleate in nature. The nucleus was very distinct, rounded in shape and regular in outline (Fig. 75 and 79).

(c) SIX DAYS OLD FEMALE FLIES OF MUSCA DOMESTICA

The flies fed with carbohydrate and lipid diets showed that the follicular epithelium was multicell layered in thickness. It was very much thickened along the entire length of the resorbed oocytes. The nuclei were pycnotic in nature and the cytoplasm was very much vacuolated and mostly found in a thin layer around the nucleus of its respective cell. The oocyte was very much reduced and irregular in size and became 1/3 in size of the whole follicle i.e. it is ready to be resorbed. The degree of vacuolization was very high in the trophocytes where the nucleolasm as well as the cytoplasm of the trophocyte were very much vacuolated. The vacuoles
were of variable sizes and were found irregularly scattered in the trophocyte cytoplasm. The trophocytes were mononucleate in nature as found in 2 days and 4 days old female flies. The nuclei of the trophocytes were very prominent and very big in size. They were irregular in outline and having variable number of nucleoli (Fig. No. 76 and 80).

(d) EIGHT DAYS OLD FEMALE FLIES OF MUSCA DOMESTICA

The flies ingested with carbohydrate and lipid diets showed that the presence of large number of degenerated, disintegrated oocytes which were going to resorbed while some follicles were empty showed the total resorption of oocyte and nurse chamber of their corresponding follicles (Fig. No. 77 and 81). Most of the follicles showed the detachment of chorion and follicular epithelium with irregular, reduced oocytes in comparison to control group (Fig. No. 77 and 81).

(C) HISTOCHEMICAL OBSERVATIONS ON THE OVARIAN TISSUE OF MUSCA DOMESTICA

The gradual histochemical changes in the ovarian tissue due to nutritional stress in experimental flies are as follows:

1. DETECTION OF PROTEIN POSITIVE MATERIAL IN THE OVARIAN TISSUE OF MUSCA DOMESTICA

(i) IN CONTROL GROUP

The ovaries of control group ingested with balanced diet showed the gradual increase in protein positive material from 2 days to 6 days old egg follicles. The oocytes showed the steadily increase in protein positive PYP bodies in ooplasm. The tunica propria of developing follicles showed high intensity of protein positive material. In nurse chamber the trophocytes showed the presence of highly protein positive nuclei. The oocyte nucleus was also highly protein positive in nature. The vitelline membrane, chorion and follicular epithelium showed intensely protein positive material. Trophocyte cytoplasm was less protein positive in comparison to trophocyte nucleus. In 6 days old flies some resorbed oocytes have seen which showed the presence of reduced quantity of protein positive material (Fig. No. 82, 83, 84).
(ii) IN EXPERIMENTAL GROUP

(a) IN TWO DAYS OLD FEMALE FLIES OF MUSCA DOMESTICA

The flies ingested with protein the ovaries showed few number of pathological oocytes. Follicular epithelium was highly protein positive. It was darkly stained with Hg B.B. The ooplasm was also highly protein positive. The trophocytes were highly protein positive. The nuclei of the trophocytes were less protein positive than the cytoplasm of their respective cells.

The nucleoplasm was vacuolated but the vacuoles were totally protein negative. The trophocyte cytoplasm was also vacuolated but the vacuoles were totally protein negative. The nucleoli were highly protein positive (Fig. No. 85).

(b) IN FOUR DAYS OLD FEMALE FLIES OF MUSCA DOMESTICA

The flies ingested with protein the ovarian tissue showed the presence of large number of resorbed oocytes. Most of the resorbed oocytes showed that the follicular epithelium became intensely protein positive in comparison with the follicular epithelium of 2 days protein ingested females. The protein positive intensity of ooplasm became decreased in comparison with 2 days protein ingested females. The oocytes became decreased in size. The oocytes were mononucleate in nature and the nucleus was less protein positive while its nucleolus highly protein positive. The boundaries of the trophocytes and their nuclei were rich in protein. The nucleoplasm and the cytoplasm of the trphocytes was intensely protein positive. The trophocyte cytoplasm was vacuolated but the vacuoles were totally protein-negative (Fig. No. 86).

(c) IN SIX DAYS OLD FEMALE FLIES OF MUSCA DOMESTICA

The flies ingested with protein the ovarian tissue showed large number of pathological oocytes. Follicular epithelium which was less protein positive. The follicular epithelial nuclei were somewhat higher protein positive than its cytoplasm. The cytoplasm was very much vacuolated. The ooplasm was less protein positive than in 4 days protein fed flies. The trophocyte nuclei and the cytoplasm were less protein-positive than in 4 days protein fed female. The trophocyte cytoplasm was very much vacuolated but the vacuoles were totally protein negative. The nucleus was less protein positive than the nucleoli of the same trophocyte. The nucleoplasm
was also vacuolated but the vacuoles were totally protein negative. The nucleoli were variable in number (Fig. No. 87).

(d) IN EIGHT DAYS OLD FEMALE FLIES OF MUSCA DMESTICA

The flies ingested with protein the ovarian architecture showed the overall decrease in protein positive material in large number of resorbed oocytes while some follicles were showed the total resorption of oocytes from their respective follicles due to high rate of resorption. Most of the oocytes became irregular in outline and also became reduced in size. Follicular epithelium and chorion were highly protein positive in nature and found detached from their corresponding oocytes (Fig. No. 88).

2. DETECTION OF PAS POSITIVE MATERIAL IN THE OVARIAN TISSUE OF MUSCA DOMESTICA

(i) IN CONTROL GROUP

The ovaries of control group ingested with balanced diet showed the gradual increase in PAS positive material from 2 days to 6 days old egg follicles. The egg follicles showed the gradual increase in PAS positive material in ooplasm of developing oocytes. The tunica propria showed highly PAS positive material in comparison to the trophocytes nuclei and trophocyte cytoplasm. Oocyte showed the gradual increase in PAS positive material. Tunica propria, follicular epithelium and chorion were less PAS positive in nature in comparison to the ooplasm of the corresponding egg follicles. Oocytes showed the gradual increase in PAS positive material from 2 days to 6 days old flies. A few oocytes became resorbed to form resorptive bodies (Fig. No. 89 to 92).

(ii) IN EXPERIMENTAL GROUPS

(a) TWO DAYS OLD FEMALE FLIES OF MUSCA DOMESTICA

The flies ingested with carbohydrate the resorbed oocytes showed that the follicular epithelium was less PAS positive in nature but it was highly PAS positive than the ooplasm of the same follicle. The ooplasm was less PAS positive in nature. The trophocytes nuclei were less PAS positive than the cytoplasm of the trophocytes. The trophocytes cytoplasm was vacuolated, but the vacuoles were totally PAS negative (Fig. No. 93).
(b) IN FOUR DAYS OLD FEMALE FLIES OF *MUSCA DOMESTICA*

The flies ingested with carbohydrate the resorbed oocytes showed that the PAS positive intensity of the follicular epithelium increased. The PAS positive intensity of ooplasm was also increases. The ooplasm was very much vacuolated but the vacuoles were totally PAS negative. The boundaries of the trophocytes were highly PAS positive but the nuclei and the cytoplasm of the trophocytes were less PAS positive (Fig. No. 94).

(c) IN SIX DAYS OLD FEMALE FLIES OF *MUSCA DOMESTICA*

The flies ingested with carbohydrate the resorbed oocytes showed that the follicular epithelium was highly PAS positive. The nuclei of the follicular epithelium was highly PAS positive than the cytoplasm of their respective cells. The ooplasm was less PAS positive than the follicular epithelium. The trophocytes were highly PAS positive in comparison with the ooplasm and less PAS positive than the follicular epithelium of the same follicle. The trophocyte nuclei and the cytoplasm were very much vacuolated but the vacuoles were totally PAS negative (Fig. No. 95).

(d) IN EIGHT DAYS OLD FEMALE FLIES OF *MUSCA DOMESTICA*

In the flies, ingested with carbohydrate the resorbed oocytes showed that the high percentage of disintegration and resorption of PAS positive material from the corresponding follicles. Some follicles were empty due to the total resorption of oocytes from their respective follicle (Fig. No. 96 and 97).

3. DETECTION OF SUDANOPHILIC MATERIAL IN THE OVARIAN TISSUE OF *MUSCA DOMESTICA*

(ii) IN CONTROL GROUP

The ovaries of control group ingested with balanced diet showed the gradual increase in sudanophilic material from 2 days to 6 days old egg follicles. Nurse chamber showed highly Sudan Black 'B' positive material in trophocyte cytoplasm while trophocyte nuclei showed the presence of large number of Sudan Black 'B' positive granules. The oocyte and follicular cell nuclei showed the presence of L₁ lipid bodies and a number of lipid spheres and spherules. Tunica propria was highly

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sudanophilic in nature. Vitelline membrane was also lipophilic in nature (Fig. No. 98, 100 and 102).

(ii) IN EXPERIMENTAL GROUPS

(a) IN TWO DAYS OLD FLIES

In the flies ingested with lipid, the ovarian architecture showed the presence of less sudanophilic material in egg follicles. Trophocyte nuclei, oocyte nucleus and follicular cells nuclei also showed a low sudanophilic intensity (Fig. No. 99).

(b) FOUR DAYS OLD FLIES

In the flies ingested with lipid, the ovarian architecture showed generally L_1 type lipid bodies in follicular epithelium and trophocytes which are lipoid in nature. Ooplasm also showed the presence of lipoid bodies, lipid spheres and lipid spherules. The tunica propria was highly sudanophilic in nature (Fig. No. 101).

(c) SIX DAYS OLD FLIES

In the flies ingested with lipid, the ovarian tissue showed the presence of large number of pathological oocytes exhibited the resorption of sudnohilic material. Most of the ovarian follicles showed the presence of intensely sudanophilic material. Tunica propria was intensely sudanophilic in nature (Fig. 103 and 104).

(d) EIGHT DAYS OLD FLIES

In the flies ingested with lipid, the ovarian architecture showed the detachment of sudanophilic follicular epithelium along with chorion from the irregular pathological oocytes. Most of the resorbed oocytes showed the active degeneration of sudanophilic material from the corresponding follicles and no follicle attain such maturity which is required for egg laying. So, neither egg laying nor corpus luteum formation has been observed in these experimental flies (Fig. No. 105 and 106).
FIG. NO. 66: Photomicrograph of transverse section of ovary of 2 days old *Musca domestica* ingested with balanced diet (Control group), showing large number of egg follicles (60X).

FIG. NO. 67: Photomicrograph of longitudinal section of ovary of 4 days old *Musca domestica* ingested with balanced diet (Control group), (60X).
FIG. NO. 68: Photomicrograph of longitudinal section of ovary of 6 days old *Musca domestica* ingested with balanced diet (Control group) (60X).

FIG. NO. 69: Photomicrograph of transverse section of ovary of 6 days old *Musca domestica* ingested with balanced diet (Control group) (60X).
FIG. NO. 70: Photomicrograph of transverse section of ovary of 2 days old *Musca domestica* ingested with protein diet, showing large number of egg follicles (60X).

FIG. NO. 71: Photomicrograph of transverse section of ovary of 4 days old *Musca domestica* ingested with protein diet, (60X).
FIG. NO. 72 : Photomicrograph of transverse section of ovary of 6 days old *Musca domestica* ingested with protein diet (60X).

FIG. NO. 73 : Photomicrograph of longitudinal section of ovary of 8 days old *Musca domestica* ingested with protein, showing some empty follicles showing the total resorption of corresponding oocytes (60X).
HAEMATOXYLIN AND EOSIN STAIN

FIG. NO. 74: Photomicrograph of transverse section of ovary of 2 days old *Musca domestica* ingested with carbohydrate diet, showing some pathological egg follicles (60X).

FIG. NO. 75: Photomicrograph of transverse section of ovary of 4 days old *Musca domestica* ingested with carbohydrate diet, showing the presence of large number of resorbed follicles (60X).
HAEMATOXYLIN AND EOSIN STAIN

FIG. NO. 76: Photomicrograph of transverse section of ovary of 6 days old *Musca domestica* ingested with carbohydrate diet (60X).

FIG. NO. 77: Photomicrograph of longitudinal section of ovary of 8 days old *Musca domestica* ingested with carbohydrate diet, showing some resorbed follicles (60X).
FIG. NO. 78 : Photomicrograph of transverse section of ovary of 2 days old *Musca domestica* ingested with lipid diet showing some degenerated follicles (60X).

FIG. NO. 79 : Photomicrograph of transverse section of ovary of 4 days old *Musca domestica* ingested with lipid diet (60X).
FIG. NO. 80: Photomicrograph of transverse section of ovary of 6 days old *Musca domestica* ingested with lipid diet showing irregular contour of all oocytes in ovarian architecture (60X).

FIG. NO. 81: Photomicrograph of transverse section of ovary of 8 days old *Musca domestica* ingested with lipid diet showing the active disintegration of oocytes while some egg follicles are empty showing the total resorption of corresponding oocytes (60X).
MERCURIC BROMOPHENOL BLUE STAIN (Hg.B.B.)

FIG. NO. 82 (A and B):

Photomicrographs of transverse section (Fig. No. 82 A) and longitudinal section (Fig. No. 82 B) of ovary of 2 days old Musca domestica ingested with balanced diet (Control group) (60X).
MERCURIC BROMOPHENOL BLUE STAIN (Hg.B.B.)

FIG. NO. 83: Photomicrograph of transverse section of ovary of 4 days old *Musca domestica* ingested with balanced diet (Control group) (60X).
Fig. No. 83
MERCURIC BROMOPHENOL BLUE STAIN (Hg.B.B.)

FIG. NO. 84 (A and B):

Photomicrographs of longitudinal section of ovary of 6 days old *Musca domestica* ingested with balanced diet (Control group) (60X).
FIG. NO. 85: Photomicrograph of transverse section of ovary of 2 days old *Musca domestica* ingested with protein diet (60X).

FIG. NO. 86: Photomicrograph of transverse section of ovary of 4 days old *Musca domestica* ingested with protein diet (60X).
MERCURIC BROMOPHENOL BLUE STAIN (Hg.B.B.)

FIG. NO. 87 (A and B):

Photomicrographs of transverse section (Fig. No. 87 A) and longitudinal section (Fig. No. 87 B) of ovary of 6 days old *Musca domestica* ingested with protein diet (60X).
MERCURIC BROMOPHENOL BLUE STAIN (Hg.B.B.)

FIG. NO. 88 (A and B):

Photomicrographs of transverse section (Fig. No. 88 A) and longitudinal section (Fig. No. 88 B) of ovary of 8 days old *Musca domestica* ingested with protein diet, showing large number of irregular, reduced sized and resorbed oocytes (60X).
PERIODIC ACID SHIFF'S REACTION (PAS)

FIG. NO. 89: Photomicrograph of transverse section of ovary of 2 days old *Musca domestica* ingested with balanced diet (Control group) (60X).

FIG. NO. 90: Photomicrograph of transverse section of ovary of 4 days old *Musca domestica* ingested with balanced diet (Control group) (60X).
PERIODIC ACID SHIFF'S REACTION (PAS)

FIG. NO. 91: Photomicrograph of longitudinal section of ovary of 6 days old *Musca domestica* ingested with balanced diet (Control group) (60X).

FIG. NO. 92: Photomicrograph of transverse section of ovary of 6 days old *Musca domestica* ingested with balanced diet (Control group) (60X).
PERIODIC ACID SHIFF'S REACTION (PAS)

FIG. NO. 93: Photomicrograph of transverse section of ovary of 2 days old *Musca domestica* ingested with carbohydrate diet, showing large number of egg follicles with differentiated oocytes (60X).

FIG. NO. 94: Photomicrograph of longitudinal section of ovary of 4 days old *Musca domestica* ingested with carbohydrate diet, showing large number of egg follicles with resorbed oocytes (60X).
PERIODIC ACID SHIFF'S REACTION (PAS)

FIG. NO. 95: Photomicrograph of longitudinal section of ovary of 6 days old *Musca domestica* ingested with carbohydrate diet, showing large number degenerated follicles (60X).

FIG. NO. 96: Photomicrograph of transverse section of ovary of 8 days old *Musca domestica* ingested with carbohydrate diet, showing some degenerated follicles (60X).
PERIODIC ACID SHIFF'S REACTION (PAS)

FIG. NO. 97: Photomicrograph of longitudinal section of ovary of 8 days old *Musca domestica* ingested with carbohydrate diet, showing the presence of large number of resorbed follicles (60X).
SUDAN BLACK 'B'

FIG. NO. 98: Photomicrograph of section of ovary of 2 days old *Musca domestica* ingested with balanced diet (Control group), showing high intensity of sudanophilic material in egg follicles (60X).

FIG. NO. 99: Photomicrograph of section of egg follicles of 2 days old *Musca domestica* ingested with lipid diet, showing intense sudanophilic egg follicles (100X).
SUDAN BLACK 'B'

FIG. NO. 100: Photomicrograph of section of egg follicle of 4 days old *Musca domestica* ingested with balanced diet (Control group), highly lipoid tunica propria and vitelline membrane while follicular epithelium showing somewhat high sudanophilic intensity (100X).

FIG. NO. 101: Photomicrograph of section of egg follicle of 4 days old *Musca domestica* ingested with lipid diet, showing less sudanophilic material in comparison to control group (100X).
FIG. NO. 102: Photomicrograph of longitudinal section of egg follicle of 6 days old *Musca domestica* ingested with balanced diet (Control group), showing high intensity of sudanophilic material (100X).

FIG. NO. 103: Photomicrograph of longitudinal section of egg follicle of 6 days old *Musca domestica* ingested with lipid diet, showing somewhat decrease in lipoid and sudanophilic material (100X).
FIG. NO. 104: Photomicrograph of section of ovary of 6 days old *Musca domestica* ingested with lipid diet (100 X).
FIG. NO. 105: Photomicrograph of section of ovary of 8 days old *Musca domestica* ingested with lipid diet, showing the resorption of sudanophilic material from large number of egg follicles (60X).

FIG. NO. 106: Photomicrograph of section of mature oocyte of 8 days old *Musca domestica* ingested with lipid diet, showing a few number of lipid bodies in the ooplasm of mature oocyte which is irregular in outline (100X).
FIG. NO. 107: Photomicrograph of section of corpus luteum of just laid *Musca domestica* ingested with balanced diet, (Control group) showing high intensity of protein (60X).

PERIODIC ACID SCHIFF'S REAGENT (PAS)

FIG. NO. 108: Photomicrograph of section of corpus luteum of 2 days laid *Musca domestica* ingested with balanced diet (Control group) showing large number of vacuoles in less PAS positive cytoplasmic syncytium. Tunica propria is highly PAS positive in nature (100X).
FIG. NO. 109: Photomicrograph of section of corpus leuteum of 6 days laid female of *Musca domestica* ingested with *balanced* diet, showing the total resorption of sudanophilic material from the cytoplasmic syncytium where hypervacuolation has seen. Tunica propria is lipoid in nature while pycnotic nuclei showed intense lipoid activity (100X).
DISCUSSION

Nutrients play very important role in the overall growth as well as reproductive potency of insects as studied in *Musca domestica* in the present investigation as reported by Taylor (1963) in housefly.

Researches have been carried out on the polytrophic ovaries, which is discoidal in shape as studied by Morrison and Davies (1964 a, b). The general anatomy of the reproductive system of the female housefly was described by Hewitt (1914) and reviewed again by West (1951). The development of ovaries was same as observed by Nicholson (1921) in *Anopheles*, Vishwanath (1924) in *Culex*, French and Garner (1965) in *Musca domestica* and Deloof et al. (1990) in *Sarcophaga bullata*. Each ovary contains about 50 to 75 ovarioles (West, 1951) and sometimes more the number depending on the conditions during larval growth on special diet (Greenberg, 1955; Taylor, 1963; Sakurai, 1977 and Trepte, 1979, 1981) as observed in the present investigation while in *Sarcophaga lineatocollis* about 25-30 ovarioles (Bhide, 1986; Bhide and Sahai, 1986). Each ovariole contains one oocyte. The penultimate egg follicle is smaller and is situated near the germarium. Three main zones are recognizable in dipteran ovariole (Lineva, 1953; Harlow, 1956; King and Devine, 1965) e.g. (i) The terminal filament – a dorsal thread like continuation often combining with others to attach the ovariole to the dorsal diaphragm by suspensory filament. (ii) The germarium – containing the primordial germ cells (oogonia) which becomes differentiated into oocyte and nurse cells and (iii) The vitellarium – comprising one to a series of follicles (egg chambers) each surrounded by a single layer of follicular cells (the follicular epithelium) which are of somatic origin. In addition there are the ovariole sheath, a thin walled tube leading to the oviduct; the follicular stalk connecting the egg chambers in an ovariole and comprising modified follicular cells (King and Devine, 1965; Goodman et al., 1968; Bhide, 1986 and Bhide and Sahai, 1986).

In the housefly three egg chambers develop simultaneously in each ovariole (Goodman et al., 1968), whereas in *Drosophila* 6 or 7 egg follicles (King et al., 1956) and in *Glossina* 2 egg follicles (Hagan 1951) develop at once in a single ovariole.
In the present investigation in *Musca domestica* the panultimate egg follicle did not grow until the first one (ultimate follicle) is fully developed and has ovulated as observed in *Sarcophaga lineatocollis* (Bhide, 1986 and Bhide and Sahai, 1986). In *Dipteran* insects in which 6-8 follicles develop together within one ovariole, yolk deposition in the ultimate oocyte (nearest to the pedicle) is restricted as in *Musca domestica* (Morrison, 1963 and Goodman et al., 1968) as also seen in the present investigation.

The development of the oocyte in *Musca domestica* could be divided into three successive phases e.g.

(i) The previtelline phase – Newly emerged to 2 days old

(ii) The vitelline phase with two subphases

  (a) The early vitelline phase – from 3\textsuperscript{rd} to 4\textsuperscript{th} days old

  (b) The late vitelline phase – from 5\textsuperscript{th} to 6\textsuperscript{th} days old

(iii) Postvitelline phase – from 7\textsuperscript{th} day onwards before oviposition.

Compared to other Diptera with polytrophic ovaries the process of oogenesis was reported by King et al.(1956); King (1960a, b); King and Devine (1965); King (1970); Cummings and King (1969); in *Drosophila melanogaster*; Fiil (1976) in *Anopheles gambiae*; Bilinski (1979) in *Campodea* Sp; Avancini and Prado (1986) in *Chrysomya putoria*; Ramadan et al. (1986) in *Culex pipiens* while in the housefly it was slower than that of *Drosophila* (King et al., 1956) yet faster than that of *Calliphora* (Thomsen, 1942) *Lucilia* (Webber, 1955); *Protophormia* (Harlow, 1956) and *Cochliomyia* (LaChance and Bruns, 1963).

In the house fly, as in *Calliphora* and *Cochliomyia*, there was a maximum of three egg chambers developing in an ovariole at one time, whereas in *Drosophila* there were six or seven. In the house fly, yolk formation was restricted to the most posterior developing oocyte in each ovariole. It was repeatedly observed (from histological sections and dissections) that all the eggs of a single ovarian cycle matured simultaneously and about 125 eggs were laid in a single batch. This characteristic of the female housefly has been termed ovarian synchrony (Morrison, 1963).
In *Musca domestica* the previtelline phase as in the newly emerged fly was the same as observed in *Drosophila melanogaster* by King and Koch (1963) is surrounded by follicular epithelium as observed by Junquera (1983) in *Dipteran* insect and the germarium contains an oogonium.

The follicle descends into the vitellarium and begins to grow as observed by Gutzeit et al. (1993) in *Apis mellifera*. After four successive mitotic divisions the oogonium is divided into 16 cells. The oocyte is differentiated sooner than the remaining 15 trophocytes, which take an active part in oocyte growth by keeping the developing oocyte continuously supplied with their contents, as is generally the case in *Muscidae* (Verhein, 1921) and *Drosophila* (King, 1966). Matuszewski (1968) studied the regulation of nurse cells in the development of egg follicle in *Cecidomyiidae* (Diptera). Oocyte is clearly differentiated from rest of the nurse cells due to its big size in comparison to nurse cells in *Musca domestica* as observed by Peacock and Gresson (1928) in *tenlivedia*, Dapples and King (1970) in *Drosophila melanogaster*; Mahowald and Stoiber (1974) in Miastor, Bilinski (1983) and Bilinski and Tylek (1987) in *Campeca* sps. The entire follicle is surrounded by firmly attached tunica propria in *Musca domestica* in present investigation as reported by Gregorio et al. (1990) in *Dermatobia hominis*.

The trophocytes are interconnected by intercellular cytoplasmic bridge as in *Drosophila* and *Calliphora* (Verhein, 1921) and *Panorpa communis* (Ramamurthy, 1964). Hagan (1951) noticed no such cytoplasmic connections in *Glossina* but they were later observed in the insect *Tsetse fly* sps. by Odhiambo (1968), Sahai (1971, 1982). in *Sarcophaga lineatocollis* observed the same cytoplasmic connections between the trophocytes and between the trophocytes and the oocyte. Ramamurthy (1984) likewise described the presence of "Fusosomes" connecting the trophocytes with one another and with the oocyte in the queen honey-bee.

Bier (1963) reported that the nurse cells in *Musca* were provided with cytoplasmic bridges between adjacent nurse cells. In *Sarcophaga lineatocollis* (Bhide, 1986; Bhide and Sahai, 1986) these cytoplasmic interconections were clearly found to be localized between the trophocytes themselves and between the trophocytes and the oocyte as also observed in *Musca domestica* in the present investigation.
In *Musca domestica* one character shared with hymenoptera follicular cells as well as with the follicular cells of snowflea (Mecoptera Boreidae) is the development of intercellular bridges arising during mitosis of follicular cells (Buning, 1993) while Bodnaryk and Morrison (1966) reported the relationship between nutrition, haemolymph proteins and ovarian development in *Musca domestica* and find out that not only the nurse cells contributed the protein, PYP bodies to the oocytes but follicular epithelial intercellular spaces also play the important role in the contribution of different metabolites from the haemolymph to the developing oocyte as observed by Dadd (1964) in Waxmoth.

The entire developing egg follicle display dense accumulation of protein ready for utilization by the developing oocyte in the early vitelline phase. The previtelline phase corresponds to the initial growth phase (Stage 1-7 or 1-8) in the House fly (Goodman et al., 1968) was also observed in the present investigation.

**Special diets** (e.g. rich in protein, carbohydrate or fat) have no effect on this phase (Previtelline phase), which is dependent on nutrient reserves such as the larval fat body in the adult insect laid down by the larva (Goodman 1963) while Derbeneva (1935) reported the influence of nutrition on the ovaries of *Musca domestica* while Ascher and Levinson (1956) studied the influence of protein addition to the larval diet on oviposition of housefly. According to Orr (1964) in blowfly *Phormia regina* Sakurai (1977) CA (Corpus allatum) in *Musca domestica* is responsible for follicle growth. Removal of CA arresting the growth of follicle. This indicates clear relationship between ovarian development and CA function. The process of vitellogenesis in insects is correlated with incorporation of yolk protein (vitellagenin) into oocyte via follicular epithelium cells (Telfer, 1965; King and Aggarwal 1965) and the fat bodies have been regarded as the most probable site of vitellogenin synthesis (Sahai, 1971, 1982; Bhide, 1978, 1986; Bhide et al., 2001) in *Sarcophaga lineatocollis* as also observed in *Musca domestica* in the present investigation.

After the previtelline phase, in 4 to 6 day old females, the availability of an adequate amount of protein in the haemolymph is the main factor in yolk deposition in the ooplasm as observed by Bownes (1980, 1982) in *Drosophila* SPS. The vitelline phase is also divided into two sub phases corresponding to stages 11 and 12.
described by Goodman et al. (1968) in the Housefly as observed in the present investigation.

As the cytoplasmic flow increases from the nurse chamber into the oocyte, the size of the nurse chamber gradually decreased, consequently the volume of the nurse chamber decreases correspond to the increases in the volume of oocyte (Trepte, 1979). In nurse chamber proteins are synthesized as long as it grows (Trepte, 1979), proteins are synthesized from the beginning to the end of the oocyte development and the oocyte reaches its final size in about 145 hrs (Trepte, 1979) as observed in *Musca domestica* in the present investigation.

In *Musca domestica* the early phase of vitellogenesis, the trophocytes grow faster than the oocyte and synthesize mRNA, which helps in protein synthesis in the trophocyte cytoplasm, the later is thus rich in protein, which the trophocyte afterwards supply to the developing oocyte. As a result of this high protein diet, the neurosecretory cells of the brain stimulate the fat bodies of the adult flies to synthesize vitellogenin, which is released in the haemolymph in the form of protein yolk precursors as observed by Bodnaryk and Morrison (1966) in *Musca domestica* and Wilkens (1968) in *Sarcophaga bullata*. The uptake of protein yolk precursors by the ooplasm, through micropinocytosis via the follicular epithelium takes place continuously from the early to the late stage of vitellogenesis as observed by Meola et al. (1977) in *Aedes aegypti*, Brennan et al. (1982) in *Drosophila melanogaster*. In the early phase of vitellogenesis the vitelline membrane is laid down as a cuticular secretion of the follicular epithelial cells. In the late phase of vitellogenesis, PYP and LYP bodies, lipid spheres and lipid granules abundant in the ooplasm Generally follicular cells are of high synthetic activity during their whole life time. The L1 bodies are the only kind of lipid bodies present in the nurse cells, the follicular epithelial cells and very young oocytes. The L1 bodies are rich in phospholipids. The L1 bodies are composed of lipoprotein and phospholipids of a very saturated nature as observed by Vishwanath (1968) in *Culex* and *Aedes aegypti* after Formaldehyde Calcium Fixatives and postchroming and nurse cells or the follicular epithelial cells never contain any L2 and L3 lipid bodies at all as observed in *Musca domestica* in the present investigation. Secretion of the chorion between the vitelline membrane
and the follicular epithelium is established in late stages of vitellogenesis as also observed in the present investigation.

The ooplasm generally contains a large number of large, centrally localized lipid spheres and a smaller number of peripherally localized smaller spheres. According to Goodman et al. (1968) the large lipid spheres in the housefly are formed by the fusion of small lipid spherules. Large quantities of lipoproteins are generally found in Musca (Verhein, 1921; and King, 1960 a, b; Goodman et al., 1968) and Glossina (Hagan 1951; Odhiambo 1968). These lipoprotein spheres are homologous with the alpha spheres of Drosophila and made up of phospholipid protein complexes (King 1960 a, b).

The possible ways in which yolk accumulates in the oocyte have been determined in many insects by Verhein (1921); Hagan (1951); Hsu (1952); Bier (1963); Kessel and Beams (1963); Morrison (1963); Coles (1964); Morrison and Davis (1964 a, b); Orr (1964); Ramamurthy (1964); King and Aggarwal (1965); Goodman (1963); Telfer (1965); Sahai (1971); Young and Hagedorn (1977) and Ramamurthy (1984). They found that protein was synthesized outside the ovary, in the fat bodies of the haemolymph, and this extra ovarian protein reached the surface of the oocyte wall and then-according to the majority entered the oocyte by micropinocytosis, while Young and Hagedorn (1977), that it entered the follicular cell interspaces and ultimately reached the oocytes. In this way, it demonstrated the dynamics of vitellogenin uptake by developing oocytes by injecting trypan blue into the haemolymph and following its course right up to the oocyte and arrived at the same conclusion as the other authors. The process of extra ovarian protein uptake in Sarcophaga lineatocollis is presumably similar to its uptake in other Diptera e.g. in housefly by Ascher and Levinsin (1956) as observed in Musca domestica in the present investigation.

Although the vitelline membrane and chorion have been formed in the late vitelline phase, they do not inhibit the passage of protein, because they are permeable for molecules of an even higher molecular weight. In Sarcophaga lineatocollis the follicular cells play an important role in secretion of the vitelline membrane and chorion as in Drosophila (King and Koch, 1963) as observed in the Musca domestica in the present investigation.
In *Sarcophaga lineatocollis* the ooplasm is packed with PYP bodies and large numbers of irregularly distributed lipid spheres and granules as observed by Hsu. (1952, 1953) in *Drosophila*, by Roth and Porter (1964) in *Aedes aegypti* and by Goodman (1963) in the house flies. Large number of PYP, yolk bodies in vicinity of the follicular epithelium, indicating that these are presumable sites of active protein synthesis in *Sarcophaga lineatocollis* (Sahai 1971).

In all developing egg follicles the tunica propria remains firmly attached to the follicular epithelium as observed by Gregorio et al. (1990) in *Dermatobia hominis*. In the early stages it is rich in protein while after the late vitelline phase it is less proteinaceous as also observed in *Musca domestica* in the present investigation.

Orientation of ovarioles and their follicles around a centre in each ovary has been reported for *Musca* (Kleine-Schonnefeld and Engels, 1981) and for *Sarcophaga* (Geysen et al., 1988). In both species follicles are oriented with their future dorsal side next to a centre, which is first indicated by the position of germinal vesicle and later on by the position of rapid building follicular cells, which lie adjacent to the germinal vesicle.

In *Musca domestica* the cross sections of ovaries of protein, carbohydrate and lipid fed female flies during chorionogenesis showed dorsal ventral polarity of oocytes by raphe structure and orientation of ovaries to an imaginary centre inside the ovary as seen in the ovaries of control flies in the present investigation as also reported by Kleine Schonnefeld and Engels (1981) in *Musca* and Geysen et al. (1988) in *Sarcophaga* respectively.

Bhide (1978, 1986) observed that maturation of the follicle in *Sarcophaga lineatocollis* was followed by ovulation as observed in *Aedes aegypti* by Curtin and Jones (1961) and ovulation was followed by the formation of yellow coloured bodies very similar to the corpora lutea of *Bombus* and *Psithyrus* (Palm, 1948) at the junction of the ovariule and the pedicle. In *Bombus* the corpus luteum is formed from nurse follicular epithelium, which absorbs the necrotic follicular epithelium of the egg, while in *Psithyrus*, in pathological cases involving degeneration and resorption of a large number of oocyte, a similar process occurs in the egg follicle, but no pigment is observed in the corpus luteum. In the Housefly (Goodman et al.,
1968; Bhide and Sahai, 1986; Bhide et al., 2001) yellow bodies are only observed after ovulation at the junction of the ovariole and the pedicle as also observed in *Musca domestica* in the present investigation.

The corpus luteum of *Sarcophaga lineatocollis* is formed as the egg is expelled from its respective follicle. The wall of the follicle gradually contracts. Early signs of degeneration are clearly discernible in the follicular epithelium whose cellular nature is completely obliterated and conspicuous changes occur in the nuclei. The chromatin material shifts at one end, the nucleoplasm contains vacuoles and the nuclei become pyknotic and are scattered about irregularly in cytoplasmic syncytia.

In the early stages of corpus luteum formation there are numerous pyknotic nuclei, but later on their number diminishes, as in the housefly (Goodman et al., 1968) and as already found in *Sarcophaga lineatocollis* by Sahai (1971, 1982) and Bhide (1978). The same observation have been noticed in *Musca domestica* in the present investigation.

In control group large number of yellow colour bodies were formed which are equal to the number of oocytes laid down by the corresponding females while in experimental group (ingested with protein, carbohydrate or lipid) due to nutritional stress the egg laying was totally arrested because the terminal follicle did not attain that maturity which is required for egg laying, so in these experimental groups due to anamalies in the respective oocytes no egg laying has seen which is due to the resorption of large number of egg follicles, so it could not be possible to locate the corpus luteum in the experimental groups in *Musca domestica* in the present investigation as observed by Galun and Fraenkel (1957) in *Aedes aegypti*, *Sarcophaga bullata* and *Musca domestica* after ingestion with carbohydrate nutrition while Goodman (1963) reported the effect of nutrition with respect to ageing in *Musca domestica*.

The tunica propria is of proteinaceous nature. Its folding and refolding is indication of active destruction and resorption of the tissue of the corpus luteum by the necrotic pyknotic nuclei in control group in *Musca domestica*. In the late stage the syncytium is highly vacuolated, indicating that the rest of the follicle is presumably destroyed. As the proceeding follicle matures, the corpus luteum
became ready for absorption, as observed by Bhide (1978, 1982); Bhide and Sahai (1986) in the same insect.

The work on histochemistry of polytrophic ovaries of dipteran insects is very scanty and has been done by Bonhag (1955 a, b) on the histochemistry of PAS+ve material in *Oncopeltus fasciatus* and *Anisolabis maritima*, King (1960 a, b, 1962, 1964); Cummings and King (1969) regarding the histochemistry and cytological studies of oocyte of *Drosophila melanogaster*, Goodman (1963), Goodman et al. (1968) on the histochemistry and cytology of ovarian development in *Musca domestica*; Galun and Frankel (1957) in *Sarcophaga bullat* and *Aedes aegypti*, fed with carbohydrate, Sahai (1971, 1975, 1982) and Bhide (1978, 1986) on the cytological and histochemical studies of *Sarcophaga lineatocollis* of normal and sugar, water fed female flies but no author has paid any attention about the depletion of metabolites in *Musca domestica* after nutritional stress as in the present investigation.

The histological and histochemical work done on the development of the egg follicle of nutritional stressed flies in *Muscidae* is very scanty and mainly concerns the housefly (Goodman et al. 1968) and *Sarcophaga lineatocollis* (Sahai 1971, 1975 and Bhide 1978, 1986).

In *Musca domestica* those oocytes which did not ovulate due to adverse physiological conditions e.g. nutritional stress. In the present investigation most of the terminal oocytes resorbed to form resorptive bodies.

If an oocyte is unable to ovulate owing to adverse physiological or environmental conditions as nutrition stress in the present investigation then instead of decaying it degenerates and is resorbed to form pigmented bodies known as "resorption bodies". In the present study oocyte resorption in the female housefly was investigated. In the early stages of resorption the follicular epithelium is normal as in control female, it consists of a single layer of distinctly circumscribed cells, each with a clearly discernible nucleus; at the distal end (away from the nurse chamber) it has several cell layers. Later on, definite sign of degeneration could be seen in the trophocytes.
In the early stages the trophocytes are mononucleate. In normally developed females the nucleus is round, while at the time of resorption its contours are irregular; the chromatin in the trophocyte nucleus becomes highly vacuolated, as a sign of active degeneration and resorption.

In control group in newly emerged female, the tunica propria is rich in protein +ve, PAS positive material, with some amount of sudanophilic material as observed in protein, carbohydrate and lipid ingested flies of *Musca domestica* in the present investigation although there is no difference between the trophocytes and the oocytes. In control group the flies aged 2, 4 and 6 days, the degree of protein +ve, PAS-positively and sudanophilic intensity of the egg follicle steadily increases while in experimental group due to depletion of metabolites there was a gradual decrease in protein +ve, PAS +ve and sudanophilic material in *Musca domestica* as observed in the present investigation but the development of the follicle is normal. In the sugar water fed females, oogenesis was somewhat slower in comparison to protein fed females. The ingestion of females with a sufficient amount of protein diet yields surprising results. Protein synthesis was seen in the trophocyte cytoplasm and also in the ooplasm. It was found that a sufficient amount of protein in the haemolymph initiated vitellogenin synthesis by the fat body and its uptake by the developing oocyte by micropinocytosis via the follicular epithelium or by the passage of vitellogenin through the follicular intercellular spaces as in *Musca domestica* (Bier, 1963); *Calliphora erythrocephala* (Thomson and Mollar, 1963); *Aedes aegypti* (Roth and Porter 1964) and *Panorpa communis* (Ramamurthy, 1964). An adequate protein diet is thus the main vitellogenin synthesis factor in *Sarcophaga lineatocollis* as observed in the housefly (Morrison, 1963; Goodman, 1963; Morrison and Davies, 1964 a, b) as also observed in *Musca domestica* after ingestion of female flies with sufficient protein diet in the present investigation, while the other experimental groups showed the presence of large number of resorbed oocytes which were irregular in outline, reduced in size with large number of variable sized vacuoles showed the site of active resorption in the present investigation as observed by Goodman et al. (1968).

Differentiation of the trophocytes and the oocyte takes place in the previtelline phase and the nurse cells then begin to grow to their full size. The nurse
cells contain highly PAS positive material in carbohydrate fed flies of *Musca domestica* as observed in the housefly (Goodman et al. 1968), and highly protein positive material with less lipoid nature in protein and lipid fed female flies of experimental group of *Musca domestica* as observed in *Sarcophaga lineatocollis* (Sahai, 1971; 1975; Bhide, 1978, 1986). Via cytoplasmic bridges, the trophocytes decant their protein positive, PAS positive and sudanophilic contents into the developing oocytes of control and experimental groups in *Musca domestica* in the present investigation.

In the vitelline phase the oocyte develops. The follicular epithelium is highly protein positive, PAS positive and sudanophilic in nature indicating that protein positive, PAS positive and sudanophilic materials are presumably absorbed from the haemolymph and passed into the developing oocyte. Towards the end of the vitelline phase the oocyte attains its full size and both the number and the size of the trophocytes steadily diminish, implying that they take part actively in oocyte development. Although the oocyte is now fully-grown and showed the presence of protein positive, PAS positive and sudanophilic material in its cytoplasm. The vitelline membrane and chorion were formed as in the housefly (Goodman et al. 1968) and as already described in *Sarcophaga lineatocollis* (Sahai, 1971; 1975, 1982; Bhide, 1978, 1986). The trophocytes of experimental groups showed the sign of degeneration and reduction as the oocyte became fully grown as also seen in control group of *Musca domestica* in the present investigation.

The strongly protein and PAS positive yolk bodies observed in *Oncopeltus fasciatus* (Bonhag, 1955a, b) and *Drosophila melanogaster* (King 1960a, b) showed that the protein is not present in a single form, but in the form of a glycoprotein or mucoprotein, as reported in *Oncopeltus fasciatus* (Bonhag, 1955a, b) and *Drosophila melanogaster* (King 1960a, b) and *Pantala flavescens* (Seshachar and Bagga, 1963) as observed in control and experimental groups of *Musca domestica* in present investigation. King and Aggarwal (1965) found glycogen deposits in the oocyte of the saturniid moth, *Hyalophora cecropia* until oogenesis was completed. Glycogen synthesis starts and the follicular epithelium begins to form the chorion (Sakurai, 1973; Trepte, 1979) points out that the glycogen content increases suddenly in the later stages of oogenesis as observed in the present investigation.
The follicular epithelium reaches its maximum volume on day 5th. Although it must be considered that the main task of the follicular epithelium is to form chorion. The investigation of Chia and Morrison (1972) indicated that the follicular epithelium obviously contributed to some extent to yolk synthesis as seen in protein fed flies in the present investigation.

In *Musca domestica* the terminal oocytes did not attain that maturity (mostly in protein, carbohydrate and lipid ingested flies) which is required for ovulation, so most of the oocytes in cross section showing degeneration, disintegration and resorption of protein positive, PAS positive and sudanophilic material not only from nurse chamber but also from the corresponding oocytes of experimental group in the present investigation. The follicular epithelium and chorion became detached from the corresponding oocytes and became irregular in outline showing protein positive and PAS positive nature.

The early sign of degeneration is vacuolation in the vicinity of the follicular epithelium; later on the entire ooplasm becomes vacuolated. The vacuoles are then larger in size and fewer in number, suggesting that the small vacuoles fused to form large vacuoles which are neither proteinaceous nor PAS positive in nature and also not containing any sudanophilic material. The trophocytes shrinks in size, as already noted in *Sarcophaga lineatocollis* (Bhide, 1978).

Early sign of degeneration are clearly visible in the follicular epithelium. Initially this develops as in the normal egg follicle, but later the nuclei became pycnotic and the cytoplasm showed hypervacuolization and all the cells in the follicular epithelium act as lecitholytic cells or vitellophages, as in *Anopheles* (Nicholson, 1921); several parasitic Hymenoptera (Weyer, 1928); *Pteromalids* (Flander, 1935); *Puppenparasiten brachymeria* (Schneider, 1941); *Bombus* (Plam, 1948); *Calliphora* (Thomson, 1952); *Mormoniella vitripennis* (Edwards, 1954); *Diadromus* (Labeyrie, 1959); *Bombus* (Medlar, 1962); *Nasonia vitripennis* (King and Richard, 1968); *Antha sexguttata* (Shrivastava, 1971) and in *Sarcophaga lineatocollis* (Sahai, 1971, 1975; Bhide, 1978, 1986).

These lecitholytic cells help in the destruction and resorption of protein +ve, PYP, PAS +ve bodies; LYP bodies, glycoprotein and mucoprotein bodies from the
ooplasm, which they decant into the haemolymph. The process is thus the exact reverse of vitellogenesis.

The ooplasm is also vacuolated and in active resorption the number of vacuoles increases. In the early stages some sudanophilic granules are found round the trophocyte nucleus which disappears completely in active resorption. The follicular epithelium, at first highly protein positive, intensely PAS positive with little sudanophilic material became less protein positive, PAS positive in nature so in later stages the PYP and LYP bodies and the sudanophilic granules are continuously resorbed by the follicular epithelium, until finally the resorptive bodies contain a few lecithotyptic cells and a little pigment as observed in *Nasonia vitripennis* (Highnam et al., 1964; Hopkins, 1964) and in *Sarcophaga lineatocollis* (Bhide, 1978, 1986).

According to the Highnam et al. (1963) oocyte resorption is correlated to the amino acid level in the haemolymph. If this falls below a certain level the developing oocyte in the ovariole is resorbed to restore the amino acid concentration in the haemolymph to the original level.

The protein yolk is the first to be dissolved and resorbed. Absorption of the lipid yolk took longer time. The lipid were absorbed but the pigment dissolved in them became more concentrated and later on it was crystallized and deposited in the neck of the ovariole together with the remains of pycnotic nuclei as observed in *Nasonia vitripennis* (Hopkins, 1964) in which a few intensely stained lipoid granules were left at the base of the ovariole i.e. between the next mature oocyte and the peduncle.

In control group in *Musca domestica* in the present investigation ovulation (hundred percent) was followed by the yellow bodies or corpus luteum formation in which protein positive, PAS positive intensity of tunica propria was steadily increases from just after laying stage to the later stages of corpus luteum formation. The pycnotic nuclei also showed the presence of high intensity of protein and PAS positive material in comparison to less protein and PAS positive intensity of cytoplasmic syncytium as observed in *Sarcophaga lineatocollis* by Bhide (1986); Bhide and Sahai (1986) and Bhide et al. (2001) so it could be concluded that diets effect the fecundity of the experimental insects as reported by Morrison and Davies (1964a, b) in houseflies.
According to Phipps (1966) in *Acrididae*, ovulation is affected by the food supply. A poor quantity of food supply or starvation reduces the rate of ovulation by increasing the rate of resorption of the terminal oocyte, is the main cause of resorption of the oocytes in *Sarcophaga lineatocollis* within a fairly short length of time. (Sahai, 1971; Bhide, 1978, 1986; Bhide and Sahai, 1986) as observed in *Musca domestica* in present investigation.

The author agrees that the ingestion of low protein, carbohydrate or lipid diets is the main cause of oocyte resorption in *Musca domestica*, in which continuous destruction and resorption of the ovarian tissue takes place. So it could be concluded from the present investigation that the balanced diet (as provided in control group) plays very important role in the development and maturation of oocytes resulted into high rate of fecundity while in only protein or carbohydrate or lipid diet ingested flies showed the partial or total arrest of development and maturity of oocytes, so they did not attain such size which is required for oviposition and in this way while partial or total arrest of oviposition resulted into poor or nil percentage of fecundity in experimental groups. And in this way control on fertility in *Musca domestica* could be achieved as observed in the present investigation.