VXI - GASTRONOMY FORMATION

1. Gastrulation

It is now well established that in ptorygote insects the material from which the inner layer is formed arises from the mid-ventral-longitudinal region of the germ band. The manner in which it is formed varies with different orders of insects. Johanssen and Butt (1941) recognize the following methods of its formation among insects.

In Diptera and Coleoptera a middle and two lateral plates are differentiated in the germ band. The middle plate sinks inwards to form a deep median furrow (gastral furrow) which becomes converted into a tube by the union of the lateral walls beneath it. The tube then flattens out to form a continuous two-layered sheet of cells. In Lepidoptera, Hymenoptera and some Orthoptera the edges of the lateral plates separate from the middle plate and then grow together beneath it, the middle plate being thus cut off from the lateral plates to form, at first, a single-layered inner germ layer. In some other forms, e.g., some Orthoptera and some Lepidoptera the inner layer is derived by active proliferation of cells from the middle part of the germ band.

In Arachnids the germ band is formed at hatching and soon after its establishment inner layer formation begins. The process of gastrulation takes place in two stages, which appear to combine the second and third methods of Johanssen and Butt cited above. These stages are as follows:
(i) differentiation of a middle and two lateral plates;
(ii) proliferation of cells from the middle plate.

The differentiation of the middle and lateral plates is initiated by the thinning out of cells in the mid-ventral longitudinal region of the germ band. As the middle plate progressively differentiates in an anterior-posterior direction it becomes separated from the two lateral plates by faint lines. In *Arina* and some other insects they are separated from each other by well defined grooves but in this sawfly they remain less distinct throughout. The lateral plates are thicker towards their dorsal edges and gradually thin out in the more lateral parts of the germ band. Causes leading to such a differentiation in the germ band could not be ascertained with certainty. It is quite probable that an early wave of proliferation might have resulted in the thinning out of cells in the mid-ventral part of the germ band as in the moth *Prodenia oriana* (Gross and Holland, 1940) and *Pieris* (Bentham, 1857). In *A. proxima* no such proliferating cells have been observed to pass down into the yolk as has been reported by the above writers.

As the middle plate differentiates a brisk activity on the part of its cells is observed which bud off a number of cells towards the inner side (yolk side) of the germ band (Plate 1, Fig. 7, and Plate 2, Fig. 10). Soon after a mass of cells is produced which projects out from the general level of the germ band. Further proliferation results in rapid increase of the inner layer (Plate 2, Fig. 11) which spreads out fan-shaped between the germ band and the yolk. The anterior pole shows
faster development than the posterior, so that the development is far advanced in this region than towards the opposite pole, where the middle plate is yet in the processes of differentiation.

Among other insects the origin of the mesoderm has been variously described. In *Pieris* (Eastham, 1927) proliferation from the mid-ventral area of the germ band has been recorded but these cells do not give rise to the elements of the inner layer. They are reported to pass down into the yolk and disintegrate there, probably serving for the liquefaction of the yolk. Thus proliferation in *Pieris* provides means to differentiate a middle plate from two lateral plates. In *A. proxima* the middle plate differentiation is only preparatory in nature, and it is the proliferation that is principally involved in the formation of the inner layer. Excepting this localized cell proliferation, there seems to be no other source from which cells of the inner layer may be derived. In *Anis* (Nelson, 1915) a portion of the median area (middle plate) of the germ band gets deeply depressed and is separated from the two lateral plates which grow together to join beneath it. The inner layer thus enclosed constitutes the future mesoderm. In the gooseberry sawfly *Pteronidea ribesii* (Shfiq, 1954) proliferation from the midventral area of the germ band has been observed and this is shown to be the only method of formation of the inner layer.

The eggs of *A. proxima* show a progressive development of the inner layer in an antero-posterior direction. In the eggs of the same age and in different regions of the same egg, the present writer has observed two distinct processes, namely
middle plate differentiation and proliferation. This, apparently, leads to the conclusion that the inner layer is formed differently in various parts of the same egg. A careful study of sectioned material, however, showed that the various processes so observed are in reality the stages of the same process. During all this development a median invagination to form the gastrular groove has not been observed.

Among the gastrular phenomenon the development of a gastrular furrow seems to be a peculiarity shared by large number of insects, viz., Chalicodoma muraria (Carrière and Bürger, 1897); Locusta migratoria (Roonwal, 1936); Calandra oryzae (Tiegs and Murray, 1933); Oncopeltus fasciatus (Butt, 1949); Pycilla perpusilla (Sander, 1956) and others. In the light of observations made on Pteronidae (Shafiq, 1954) and the present sawfly, it may be pointed out that proliferation is the principal method of formation of the inner layer amongst sawflies. Although, the median plate in A. proxima only demarcates a certain proliferating region from the rest of the germ band, yet its existence brings this sawfly closer to Apis (Nelson, 1915) in matters of gastrulation.
2. Development of the Mesoderm upto the formation of Mesodermal Tubes

Only the origin of the mesoderm has been considered so far. In the following pages further development of this layer upto the formation of paired mesodermal tubes will be described.

The cells of the mesoderm that are proliferated either singly or in groups of two or more run into each other and begin to spread over along the inner surface of the outer layer, eventually forming a complete lining. The entire mesoderm in this manner forms a continuous mass of cells extending posteriorly from the labral region upto the last abdominal segment (Plate 7, Fig. 49). In early stages the mesodermal cells grow round the base of the stomodeal invagination so that the pre- and post-oral mesoderm becomes continuous. As the invagination further deepens the mesodermal layer does not keep pace with it and flanks the stomodeum laterally. At about 18-hours a shallow median depression appears in the outer surface of the ectoderm which forms the neural groove. Above the neural groove lies the single-layered median mesoderm from which blood cells are produced. In the more lateral parts, the mesoderm forms two or three layers. Following external segmentation of the embryo, segmental mesoderm becomes much thickened while the intersegmental parts remain narrower (Plate 2, Figs. 13 and 15). Except this piling up of cells in the segmental regions and a relatively thin mesoderm in the intersegments no well defined segmentation of the inner layer has been observed in Athelia proxima.
A similar condition has been reported by Shafiq (1954) in Pteronidae and by Nelson (1915) in the honey bee. As the buds of the appendages begin to grow in their respective segments the mesoderm extends into their developing cavities.

**The Mesodermal Tubes.** During the period in which cells are spreading out laterally (20-22-hours) a rounded cell mass appears along the lateral crests of the mesoderm on either side of the body (Plate 2, Figs. 11 and 12). These cell clumps are the rudiments of the mesodermal tubes and are found both in the segmental and intersegmental regions. They form a continuous longitudinal solid band of cells which extend from the second maxillary up to the last abdominal segment. In a transverse section each cell clump presents a rounded form, recalling the condition found in the extreme lateral parts of the mesoderm in the segmental regions of *Piersis* (Eastham, 1930). In *A. proxima* the cells are closely packed in this region but up to this stage there is no sign of a cavity. An hour or two later, a small circular cavity appears (Plate 2, Fig. 12 b, Coel) which gradually grows into a large slit-like space forming the embryonic coelom (Fig. 13).

The development of coelomic cavity among insects usually occurs in the following two ways, as illustrated by *Locusta* and *Calandra*. In *Locusta*, unlike *A. proxima*, the lateral mesoderm is divided up into segmental groups forming segmental somites. The free lateral edges of the segmental mesoderm in each lateral half become folded over to enclose the coelomic cavity. This type of development, according to Roonwal, (1937) holds true for the head,
thorax and the first abdominal segment. In the remaining trunk segments the cavity appears as a fissure in the double-layered mesoderm. In Calandra a cleft appears between the somatic and splanchnic layers of the segmental somites which forms the coelomic cavity.

In A. proxima it appears quite probable that the cells loosen and then migrate peripherally within the cell clump leaving a small circular cavity in the middle. Embryonic coelom so formed gradually grows into a large slit-like space. It may be pointed out that there is no sequence in the order of formation of these cavities. Sporadic appearance of small cavities in various regions of the cell clumps and the coalescence of the adjacent cavities eventually results in the formation of two continuous tubes running along the sides of the body. These are the mesodermal tubes (Plate 2, Fig. 14, mes) which by thirtieth hour attain their maximum development. They are connected with each other throughout their extent by the underlying main bulk of the mesoderm both in the segmental and intersegmental regions.

Among the highly specialised insects, e.g., Hymenoptera, segmental coelomic sacs like those of Locusta do not occur. They are typically developed in Orthoptera and the lower pterygote orders. In the representatives of Hymenoptera the lateral mesoderm extends longitudinally as a continuous layer and the coelomic cavity is fused in a longitudinal direction. It is restricted to the latero-dorsal part of the developing embryo. These structures have been termed as 'mesoderm röhre' by Carrière and Bürger (1897) in Chalicodoma, and the term has since been retained by subsequent workers in which they
closely resemble in fate and form. In Chalicodoma, in addition to the mesodermal tubes, paired segmental coelomic sacs have also been recorded. Carrière and Bürger (1897) state:

"... Sie sind durch querräume in Abschnitte abgetheilt, von denen je einer zu einem Mesodermsacke gehört. Die Querräume befinden sich stets in der mitte zwischen zwei mesodermsäcken."

These mesodermal sacs are dorso-ventrally flattened and are situated mesally to the mesodermal tubes. Nelson (1915) quoting Bürger (1897) says that the mesodermal sacs communicate intersegmentally with the mesodermal tubes of the corresponding side. Moreover, these tubes in Chalicodoma are divided by thin partitions in the intersegmental regions. No structures comparable to these mesodermal sacs are present in A. proxima. The mesodermal tubes of Apis (Nelson, 1915) extend from the middle of the second maxillary up to the eleventh trunk segment and closely resemble those of Athalia and Pteronidea (Shafiq, 1954) in structure. In transverse sections it appears that the lumen of the tube is narrower in the intersegmental than in the segmental regions. A similar condition has been reported by Nelson (1915) in the honey bee and by Shafiq (1954) in Pteronidea.

With the development of the mesodermal tubes the mesoderm becomes well defined. Though there is no distinction yet between the various component parts of the mesodermal layer, the terminology of Eastham (1930) has been adopted in the following account for descriptive convenience. The somitic, or that part of the mesoderm which forms the mesodermal tubes, lies towards the dorso-lateral end of the embryo. Between it and the median
mesoderm is the somitic which, as already stated, extends into the cavities of the developing appendages (Plate 2, Fig. 13, splm). In the segmental regions, particularly of the thorax, it is several layers in thickness. The median mesoderm remains single-layered throughout. The somitic mesoderm may further be differentiated into an outer thick somatic and a relatively thin, inner visceral layer of the mesodermal tube (Plate 2, Fig. 13), both of which are continuous and enclose the coelomic cavity. From the dorsal portion of the visceral wall arise the rudiments of the enteric muscles. In the beginning these are in the form of a mesally directed ridge (Plate 2, Fig. 13, splm) on either side of the body, but as they grow ventrally and dorsally they unite to form the muscular coat of the intestine (Plate 3, Fig. 22 and Plate 5, Fig. 32 splm). This condition is very similar to what has been described in Anis (Nelson, 1915). With the development of the splanchnic ridge the visceral wall is reduced to a thin layer of cells. This portion, as will be seen later, becomes closely associated with the germ cells to form the genital ridges. The mesally directed ridge sometimes, in sections, gives an impression that the free lateral edges of the mesoderm are folded over to enclose the coelomic cavity and may lead to an erroneous conclusion that the coelomic cavity originates by folding over as in Locusta (Roomval, 1937) and Rhynchoidea (Shafiq, 1954). Studies of the early stages clearly indicate that this is not possible since the coelomic cavity appears before the differentiation of the splanchnic ridge.

Contemporaneous with the development of the splanchnic layer the yolk is withdrawn from the median and lateral parts of the embryo.
The epineural sinus so formed (Plate 3, Fig. 22, Eps) becomes continuous with the coelomic cavity to give rise to the definitive body-cavity. Another change that has occurred during this part of the development is the precocious derangement of the mesodermal tubes in one or two anterior segments. The tubes now extend from the third thoracic to the last abdominal segment.
3. Mesoderm of the Head Region.

In the head region of *A. proxima* the mesoderm can be followed as a continuous layer up to the labral segment. Along the sides of the head, particularly at the bases of the appendages, the mesoderm becomes thick and extends into the developing appendicular cavities. In the middle of the head the cells are scanty and are loosely spread over to form scattered masses. In the labrum it is represented by a bilobate cell-mass, which is strongly suggestive of its paired origin. Coelomic sacs are not formed in the head region of *Athalia* but the antennal segment is a notable exception. **Antennary Coelomic sacs** The mesoderm of the antennary segment provides special interest as it gives rise to a pair of coelomic sacs (Plate 7, Fig. 50, ante). The cells of the mesoderm before the shortening of the embryo begins, are spread over irregularly but at the age of 30-hours they arrange to form a pair of coelomic sacs. These sacs lie under the brain and behind the bases of the antennal rudiments on either side of the stomodaeum. Their median walls are thicker than the outer ones (Plate 4, Fig. 25). The thin walled coelomic sacs enclose the coelomic cavity which does not extend into the hollow of the appendages. The antennary coelomic sacs of *Athalia* correspond in position to those of *Aris* (Nelson, 1915) and *Calandra* (Tiegs and Murray, 1938), but differ from the former in the absence of a mesodermal core going into the cavity of the antennal appendages.

Antennary coelomic sacs have been recorded in a number of insects. In Hymenoptera they are stated to occur in *Aris* (Nelson, 1915).
and Chalicodoma (Carrière and Bürger, 1897). Shafiq (1954) has not described them in Pteronidea, in which case they probably escaped the observations of the writer. In the majority of forms where antennary coelomic sacs have been observed, they are shown to give rise to the cephalic aorta. In Athalia proxima, in addition to giving origin to the aorta and the antennary muscles, the antennary mesoderm forms an investment of the pharyngeal ganglia and the corpora allata. This is very similar to what has been described in Locusta (Roomwal, 1937).

Posteriorly, the antennary mesoderm is connected with the mesoderm of the following segments, and from the second maxillary up to the last abdominal segment it forms the lateral mesodermal tubes. The general disposition of the mesoderm in the head region agrees in all essentials with the observations made on Apis (Nelson, 1915).

Subsequent changes in the mesoderm are principally concerned with the organogenesis and shall be described under appropriate headings in the following chapters.