

nicum (Faust and Meleney, pp. 67, 103), these larvae of *S. mansoni* may have fed outside the portal stream. Later cumulative evidence favored another view, namely, that fed larvae found in the right and left hearts, the lungs and the pleural cavity, had all been to the liver at least once and had fed there, after which they had passed through the capillary filter bed of the liver back into the right heart, lungs and left heart, most of them eventually returning to the liver. Moreover, larvae in the right heart, like those in the liver, bore evidence of having fed very recently, as indicated by the reddish-black color of their guts and the presence of undigested red blood corpuscles; while some of those in the lungs, and all in the pleural cavity and left heart, had light brown-colored guts, with no undigested erythrocytes. In addition to this evidence, the continued delay in the accumulation of the young worms in the portal stream, as contrasted to the "lag" within the respiratory circulation, differed from the condition which obtained in Faust and Meleney's experimental work (Table 1) on *Schistosoma japonicum* during the migration of the metacercaria through the tissues. However, these investigators⁴ did occasionally find in lung concentrates black-gutted larvae (p. 103), which they interpreted as having escaped from the portal circulation.

Although a few larvae were found in washings from the pleural cavity, the mediastinum and diaphragm were consistently negative for larvae or any indications of larvae having attempted to penetrate these membranes. Furthermore, there was practically no evidence in our animals of petechial hemorrhage in the stomach wall and kidneys, such as Faust and Meleney (pp. 107-111) found so characteristic of the migration phase of *S. japonicum* from the lungs to the liver.

One rabbit was sacrificed on the tenth day after inoculation. Autopsy was carried out as for the rats. Unfed larvae were recovered from the right heart and lungs, while the majority of those from perfused liver were more robust and contained red blood cells in their intestines. There was definite evidence of fresh petechiate hemorrhage in both lungs, particularly on the diaphragmatic and mediastinal aspects. The pleural cavity and diaphragm were negative.

Migration of the adolescent worms out of the liver

Up to and including the 450th hour all young worms found in the abdominal viscera were obtained from the intrahepatic portion of the portal blood vessels. About four days later (546 hours) the first evidence of migration into the mesenteric veins was obtained. In the rat sacrificed at this period many immature worms were recovered from perfusion of the liver and later from minced liver concentrates. No worms were recovered from the thoracic viscera. Careful examination of the mesenteric radicles yielded three young worms, about half-grown (*i. e. mu, nu and xi stages*). Two worms were observed in the ileocolic branch of the superior mesenteric vein (terminal ileum) and one, in the anastomosing arch between this vein and the right colic vein (middle cecum). From this time on there was continued migration of the worms out of the liver into the mesenteric venules. By the 720th hour (rat and rabbit) a considerable number of adolescent worms (none mated), but no adult forms, were obtained from the posterior branches of the superior mesenteric vein (in the region of the ileocecal junction). Even by the 40th day after inoculation no sexually mature female worms (*i. e. ovipositing females*) were recovered in the murine and rabbit hosts. In these animals a few almost mature schistosomes, together with many adolescents and some juveniles, were obtained from the liver. The specimens located in the mesenteric veins were all adolescents or almost mature males and females (*rho and sigma stages*). These were located in the ileocolic branch of the superior mesenteric vein, in the right and middle colic branches of the superior mesenteric vein, and in the left colic and sigmoid branches of the inferior mesenteric vein. By the 47th day mature worms *in copula* and eggs were found in the tissues adjacent to the right and mid colic veins of the superior mesenteric stem (Table 1). Autopsies later than this period were confined to rabbits and monkeys, which were sacrificed between the 55th and 70th days, and after 6 and 9 months. In these animals the schistosomes were, with very few exceptions, fully mature at the time of autopsy. Many more worms were found in the mesenteric veins than in the liver (about 5 to 1 in both species of hosts). Furthermore, the distribution of these worms had spread throughout the entire

mesenteric systemic and its posterior anastomoses, although the great majority were massed in the vicinity of the ileocecal junction or below this level. Representative distributions were found as follows: (1) monkey (55 days), anterior branches of superior mesentery vein, 75; ileocolic branch, 20; right and mid-colic branches, 69; left colic and sigmoid branches of inferior mesenteric vein, 151; rectal radicles of superior and middle hemorrhoidal veins, 41; main stem of mesenteric vein, 86; total yield of extra-hepatic vessels, 442, (2) rabbit (60 days), anterior branches of superior mesenteric vein, 49; ileocolic branch, 378; right and mid-colic branches, 39; left colic and sigmoid branches of inferior mesenteric vein, 82; inferior hemorrhoidal vein and vesical plexus, 8; main stem of mesenteric vein, 200; total yield of mesenteric veins and collateral vessels, 756; (3) monkey (70 days), anterior branches of superior mesenteric vein, 7; ileocolic branch, 10; right and mid-colic branches, 62; left colic branch, 2; hemorrhoidals, 0; total yield of mesenteric veins, 81; (4) rabbit (9 months), anterior branches of superior mesenteric vein, 117; ileocolic branch, 597; right and mid-colic branches, 114; left colic branch, 164; hemorrhoidals and vesical plexus, 314; main stem of mesenteric vein, 0; total yield of mesenteric veins and collateral circulation, 1,306.

Analysis of these findings provides a basis for the following statements. In *Schistosoma mansoni* the worms, while still frankly immature (stages *iota* to *kappa*), begin to migrate out of the liver into the mesenteric veins. Our data indicate that in the rat this migration begins about the 23rd day after inoculation and only a few days after the worms had definitely accumulated in the intra-hepatic portal stream. The earliest immigrants, like most of the later ones, have a primary predilection for the veins draining the region of the bowel in the vicinity of the ileocecal junction. Later stages of this infection have a tendency, probably because of crowding, to spread along the entire mesenteric system and even to invade the hemorrhoidal vessels and vesical plexus, although their greatest concentration is in the vicinity of the cecum. At least up to the 9th month after inoculation the worms in the mesenteric vessels, on the average, outnumber those within the liver about five to one.

In a preliminary series of experimental mammalian infections with *Schistosoma mansoni*, in which rats, guinea pigs

and monkeys were used, at autopsy of the hosts, adolescent and mature worms, as well as eggs, were found in the lungs. These worms were invariably in pulmonary arterioles, were mostly adolescent or slightly immature even three or six months after inoculation, and were seldom paired. In the rabbit sacrificed at the end of the 9th month numerous slightly immature degenerating *S. mansoni* were found. They were invariably surrounded by neutrophiles, eosinophiles and macrophages, and in some instances the immediate areas had undergone partial fibrosis. Although these findings of *S. mansoni* in the lungs have been previously duplicated in post-mortem cases of *Schistosoma haematobium*, *S. mansoni* and *S. japonicum* infection, they require an explanation.

Our studies have indicated that growth and development of the schistosomula beyond the very earliest post-cercarial stages can take place only after the young worms have been in the portal blood stream. It seems reasonable to believe that maturing worms in the pulmonary arterioles had passed their juvenile period in the portal vessels within the liver, had later migrated out into the inferior mesenteric veins, had passed into the median hemorrhoidal vessel and had been carried into the right heart through the inferior vena cava. From the right heart they had passed into the pulmonary circulation, where they became lodged in the arterioles and continued a slow, subnormal development, considerably behind that of worms in the more normal habitat of the mesenteric radicles and their immediate anastomoses. Some of these worms in the lungs probably never reach maturity, but are attacked by tissue elements and die in the adolescent stage. This view is supported by the following evidence: In rabbits and monkeys sacrificed after the beginning of oviposition, some worms were usually found in the hemorrhoidal veins. In one rabbit (45 days after inoculation) five males and two females were recovered from the superior and middle hemorrhoidal veins. In another (sacrificed on the 60th day after exposure), 46 males and 30 females were found in the superior and middle hemorrhoidal veins and four pairs in the inferior hemorrhoidal veins and vesical plexus. In a monkey, sacrificed on the 55th day after inoculation, 24 males and 17 females were recovered from the superior and middle hemorrhoidal vessels and one mature pair was found in a clot of blood in the main postcaval vein. In one rabbit, autopsied eight

months after inoculation, three males and two females were recovered from the vesical plexus. These findings indicate that in moderate to heavy infections of the experimental host a small proportion of the worms becomes located in vessels within relatively ready access to the right heart. Furthermore, we have a section of bladder from a human autopsy in which many hundreds of *Schistosoma mansoni* eggs are infiltrated through the wall. It was the occasional discovery of lateral-spined schistosome eggs in the urine and their presence in the bladder wall, together with similar findings of *S. haematobium* eggs in the intestinal wall, which was responsible in no small measure for the confusion regarding these two species in Egypt.

The successive stages of development of Schistosoma mansoni in the mammalian body and their time span

(Chart 1)

When the cercariae of *Schistosoma mansoni* enter the skin, they leave their tails behind, and, with the caudal trunk, the posteriormost pair of flame-cells (solenocytes). Thus the first stage in the mammalian body (the *alpha* stage) possesses only three pairs of flame-cells. The head organ is still proboscis-like, with the minute opening of the digestive tract situated midventrally some little distance behind the anterior extremity. By the time the larva has penetrated through the epidermal layers of the skin it has used up practically all of its penetration gland contents, so that these organs are very much shrunken, and consequently have very little lytic material to aid in proceeding actively through the tissues from this time on. There is abundant evidence, however, that they do reach the peripheral venules, are able to work their way through lymph nodes in which they may be caught, and, slightly later, may rupture pulmonary capillaries. It is altogether likely that this progress is accomplished primarily by the mechanical movements of the larva, which "noses" its way with its very active proboscis through loose tissues and into small vessels. When the larva has reached the *rete mucosum* it still maintains its essential size and shape as a decaudated cercaria, with transverse diameters of 50×35 microns (Fig. 1). Its capacity for elongation and, at the same time, diminution of its diameter,

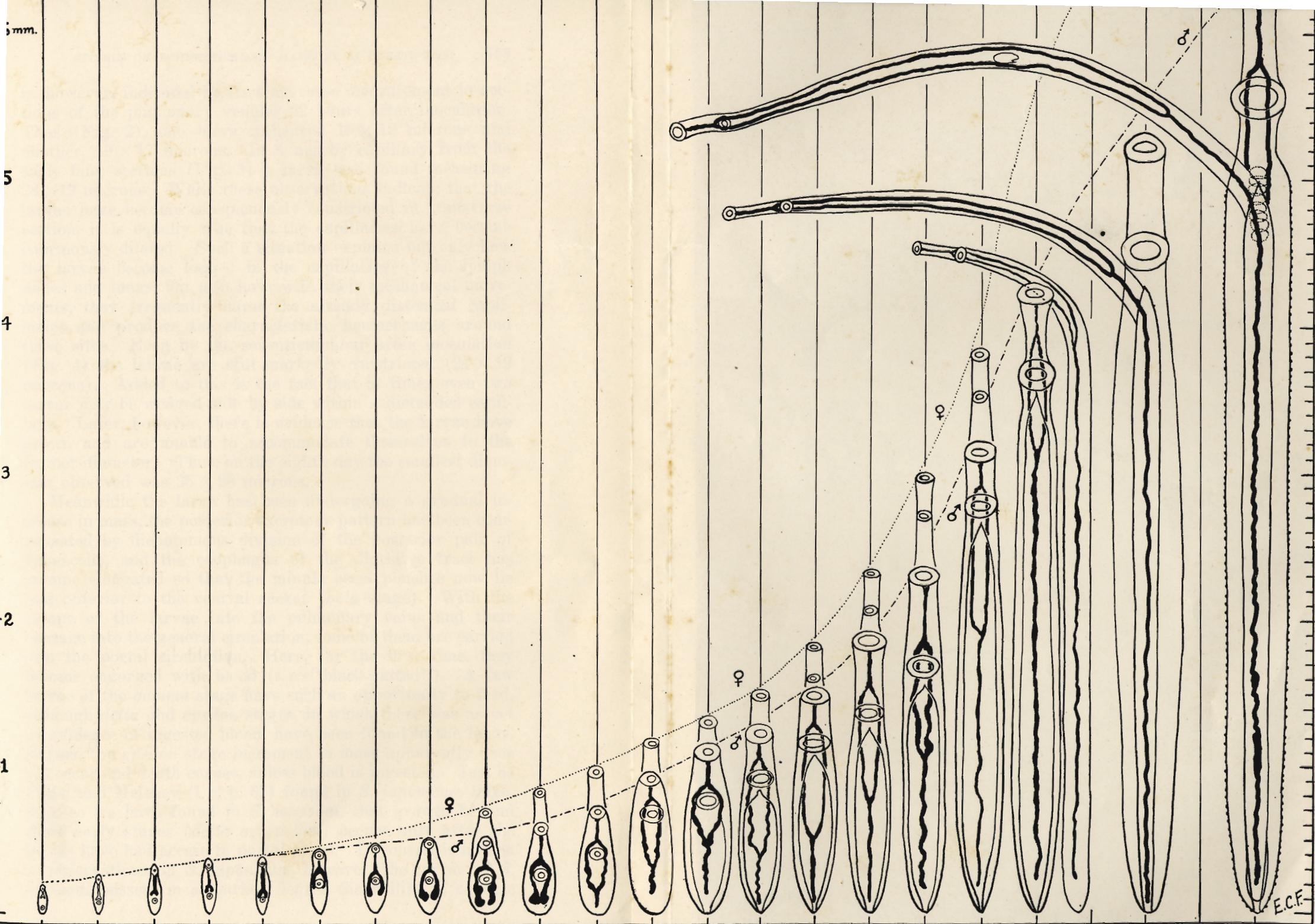


CHART 1.—Semidiagrammatic representation of the successive stages of growth

DIAGRAMA No. 1.—Representación aproximada de las etapas sucesivas de desarrollo

is, however, indicated by its transverse measurement in sections of the pulmonary venules 22 hours after inoculation. Thus (Fig. 2), one larva measured 19×12 microns and another, 19×17 microns. In a nearby capillary from the same lung sections (Fig. 3) a larva was found measuring 28×19 microns. While these observations indicate that the larvae have become conspicuously constricted in transverse section, it is equally true that the capillaries have become enormously dilated. Such a situation explains not only how the larvae become lodged in the capillaries of the lymph nodes and lungs, but also how, with their mechanical movements, they frequently burst the already distended capillaries and produce the characteristic hemorrhages around these sites. Even by the seventieth hour after inoculation (Fig. 4) the larvae are still markedly constricted (24×19 microns). Added to this is the fact that at times even two larvae may be wedged side by side within a distended capillary. Later, however, there is evidence that the larvae have grown and are unable to accommodate themselves to the former diameter. Thus, on the eighth day the smallest diameter observed was 35×28 microns.

Meanwhile the larva has been undergoing a gradual increase in mass, the posterior excretory pattern has been compensated by dichotomous division of the posterior pair of flame-cells, and the esophagus of the digestive tract has become elongated so that the minute cecal pouches now lie just anterior to the ventral sucker (*beta* stage). With the escape of the larvae into the pulmonary veins and their passage into the general circulation, some of them are carried into the portal circulation. Here, for the first time, they become engorged with blood (*i. e.* "black-gutted"). A few larvae of the *gamma* stage have such an opportunity to feed, although *delta* and *epsilon* stages, in which there was as yet no evidence of ingested blood, have been found in the lungs. Beyond the *epsilon* stage increment in mass apparently does not occur and death ensues, unless blood is ingested. Just as Faust and Meleney (*l. c.* p. 67) found in *S. japonicum* infection, so we have found in *S. mansoni*, that growth beyond these early stages (*delta* or *epsilon*) occurs only after the larvae have had access to portal blood. In contradistinction to observations on *S. japonicum*, however, the larvae of *S. mansoni* possess in a marked degree the ability to squeeze

through the portal capillary filter and other capillary networks, such as those of the stomach wall and kidneys, and to return to the lungs. Many of these larvae, when recovered from the lungs, were robust and were consistently "black-gutted" or bore evidence of having fed recently on blood.

The effect on the larva of feeding on portal blood has been adequately demonstrated. Not only is the animal more robust and its mass greater, but the intestine increases in length to accommodate more ingested blood. Furthermore, by the *zeta* stage (10 days), the proboscideal or labial organ has metamorphosed into a definite sucker, the intestinal ceca have increased in diameter and are now beginning to proceed posteriad around the ventral sucker, and the region of the body behind the ventral sucker is now increasing in width and length. Likewise, about this time, there is the first indication of sexual differentiation. The larvae which are to become males are recognized as appreciably broader and shorter than the potential females. From this time on it is possible to differentiate the two sexes.

In both male and female schistosomula the progressive changes, which have been mentioned, continue. By the *kappa* stage the intestinal ceca have approximated one another in the midline some distance behind the ventral sucker. Before the *lambda* stage they have fused at this site and subsequent stages show a continuously elongating common median posterior prolongation of the gut. The most important differential sexual development at the end of this juvenile period is the origin of the gynecophoral canal in the male. We have observed it for the first time in the *nu* stage. The broad lateral fields of the male worm, posterior to the ventral sucker, are first flattened out and later infolded ventrally toward the midline. At first (*nu* stage-) only the lateral margins begin to curve ventrally, but by the next (*xi*) stage a definite trough has been formed. From this period it seems appropriate to refer to these differentiating schistosomula as *adolescent worms*.

The adolescent period is marked by very rapid growth, which begins immediately with the formation of the gynecophoral canal of the male worm. Most of this increase is in length. At first the increment in the female is proportional to that of the male, but toward the end of the period (*tau-ypsilon* stages) the elongation of the female becomes much

more rapid. Although the worms are structurally adapted for pairing during the entire adolescent period, mating has not been observed before the *phi* stage, which is practically the end of this period.

The adult worms.—Adult worms are those which have mature functioning sex organs. In our series of experiments, in which the worms were allowed to reach maturity before sacrificing the hosts, precautions had been taken to guarantee the presence of both sexes by pooling the cercariae from several snails (*vide* Cort⁸, Faust⁹). Therefore in all of our autopsies, even when only a few worms were recovered, both males and females were present. Nevertheless, the males consistently outnumbered the females (rabbit no. 1, 346 males to 119 females; rabbit no. 2, 918 males to 503 females; rabbit no. 3, 477 males to 169 females; rabbit no. 4, 349 males to 283 females; monkey no. 1, 62 males to 38 females, and monkey no. 2, 292 males to 207 females). In monkey no. 8, sacrificed more than two years after exposure, in addition to 10 pairs, 27 males and 13 females were found. Although our experimental series comprised both those infections which had only recently matured, as well as semi-chronic and chronic types, there was no essential change in the relatively greater number of males as the infections aged. More interesting was the observation that, in all of these infections, only a small proportion of the worms was coupled, even in the terminal venules adjacent to the lower bowel wall. Nevertheless most of the uncoupled females were producing eggs. In the mated forms some 12 per cent of the females were lying in reverse position with respect to the males (*i. e.* with their heads projecting out of the distal portion of the gynecophoral canal and their posterior extremities lying near the heads of the males. In all cases, however, the heads of the females were directed toward the terminus of the venules. Taken as a whole, it appeared that there was considerable movement on the part of the worms from one site to another within the vessels, and that mating was a relatively temporary phenomenon for the females.

Leiper¹ first recovered *Schistosoma mansoni* eggs from the dejecta of the Indian monkey 42 days after inoculation. In five experimental rhesus monkeys, we first recovered eggs from the stools between the 37th and 44th days, following three or four days of prodromatal diarrhea. In an assistant,

who accidentally infected himself, eggs first appeared on the 42nd day. In our rabbits eggs were never recovered from the stools, and scrapings of the rectal mucosa between the 40th and 50th days were negative. Our sections of the large bowel of these animals indicated that the worms rarely migrated into the terminal venules and that eggs rarely infiltrated into the bowel wall.

Mature male worms (Fig. 5) are covered with sessile papillae from the level just behind the ventral sucker to the distal extremity. These papillae are of equal size and are about equally distributed over the dorsal and lateral aspects of the body, but tend to be suppressed within the gynecophoral canal. These papillae possibly help to anchor the worms in small veins. The entire integument is studded with minute thorn-like spines. The posture of the male, whether single or mated, is characterized by a marked ventral curvature of the entire body posterior to the ventral sucker, and a distinct dorsal flexure of the more constricted "neck" region.

The adult male of *S. mansoni* in our experimental rabbits and monkeys varied in size from 6.4 to 9.9 mm. in length by a greatest transverse diameter (just behind the testes) of 0.36 to 0.58 mm. This latter measurement was made with the folds of the gynecophoral canal in their functional position. Manson-Bahr¹⁹ (p. 689) states that the male's length lies between 10 and 12 mm. If these latter figures refer to worms from human infections, it might indicate that the males attain a greater size in the human host. In our material the oral sucker measured from 0.18 to 0.28 mm. in diameter and the ventral sucker, 0.28 to 0.33 mm. There was a distance of 0.5 to 0.6 mm. between the centers of these two acetabula.

The internal anatomy of the male worms was studied by us in *toto* mounts and in sections. The intestinal tract lacks a muscular pharynx, which is replaced by an enveloping cluster of glandular cells. It bifurcates just in front of the ventral sucker. The furci join again at various distances in the middle third of the body and proceed as a single blind cecum to the subdistal region of the body. The male genitalia (Fig. 6) are located near the mid sagittal plane just behind the ventral sucker. There are 6 (?) to 9 subglobose testes (usually 8), more or less alternating in position toward the dorsal aspect of the worm. From each there emerges a short

vas efferens (*VE*). The vasa efferentia open into the common vas deferens (*VD*), which is situated in the median sagittal plane ventral to the testes, and opens into the flask-shaped seminal vesicle (*VS*) on the postero-ventral aspect. A curved, non-muscular cirrus tube (*CT*) arises from the postero-dextral aspect of the seminal vesicle, passes ventrad on the right side of the vas deferens, and opens medially on the venter of the worm, within the anterior portion of the gynecophoral canal (δ *G P*). It is devoid of muscular elements. Discharge of the spermatozoa can apparently occur only by the contraction of the weakly muscular seminal vesicle. Except for the different number of testes and the more posterior opening of the vas deferens into the seminal vesicle, the genital organs in the male *S. mansoni* closely resemble those of *S. japonicum* (Severinghaus²⁰).

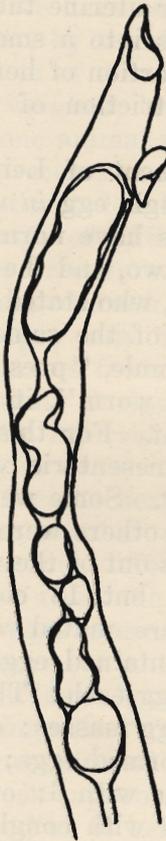
In general, female and male worms, whether coupled or alone, were found in about equal numbers in the terminal veins of our monkeys. The maturity of females in all of our animals was practically synchronous with that of the males. The mature female in our experimental rabbits and monkeys had a variation in length from 7.2 to 14 mm., and a maximum width of 0.20 to 0.38 mm. The length measurements are considerably smaller than those given by Manson-Bahr (l. c. p. 690), which range from 12 to 16 mm. These worms have no integumentary papillae but are completely covered with many minute, very sharp, thorn-like spines. The oral sucker in our material measured 0.065 to 0.075 mm. in diameter, and the ventral sucker, 0.078 to 0.085 mm. in diameter. The two acetabula were situated about 0.35 mm. apart. The intestinal tract resembles that of the male, except for its more delicate structure. The gut bifurcates just anterior to the ventral sucker. The furci proceed posteriad in the lateral fields and join mesad just behind the posterior end of the ovary. The single cecum then proceeds to the subdistal region of the worm, where it ends blindly. As in the male worm, the gut is usually filled with dark brown material which is the remains of ingested blood.

The genital system of the female worm is relatively complicated (Fig. 7). In the posterior half of the worm, from the gut junction to the subdistal region, somewhat lateral to the mid-line, are the vitellaria (*Vi*), which are coarsely granular ovoid organs, with short minute ducts connecting

mesad with the common vitelline duct. This duct (*ViD*) proceeds anteriorly across the ventral face of the ovary, and, at a distance approximately the length of the ovary anterior to that organ, joins the oviduct to empty into the oötype (*Oo*). The ovary (*O*) is an elongated oval, slightly torted organ, lying in the mid-line just anterior to the posterior junction of the intestinal furci. It is filled with many mature and developing egg cells. Its anterior end is blind. At its posterior end there arises the oviduct. Soon after its emergence posteriorly from the ovary, it gives off a crooked blind cecum, the seminal receptacle (*RS*), which is slightly more dilated at the blind end. The oviduct (*OD*) proceeds around and forward across the dorsal face of the ovary, joining the common vitelline duct before entering the oötype. The presence and position of the seminal receptacle in *S. mansoni* closely resemble that described by Severinghaus²⁰ for *S. japonicum*, except that in *S. mansoni* there apparently is no dilated fertilization chamber where this organ joins the oviduct. The oötype (*Oo*) is a short narrow chamber surrounded by a cluster of elongate glands. Immediately anterior to this site is the proximal end of the uterus (*U*). It is usually greatly dilated at this end and contains the single egg (*E*) characteristic of most *S. mansoni* females. Multiple egg groups (Faust and Hoffman¹⁰, fig. 1) or egg agglomerates (text fig. 3) are usually found farther up the uterus. The uterus is relatively thick-walled and muscular. It proceeds anteriorly, in the mid-plane between the intestinal furci, and ends in a slightly thickened enlargement just behind the ventral sucker (Fig. 8). At this site a short duct bends ventrad and provides a poorly circumscribed non-muscular vent for the eggs (♀ GP).

Actual insemination of the females has not been observed. With the poorly developed, non-muscular cirrus tube and the unprotected uterine pore, it is, indeed remarkable that fertilized eggs are so common. In coupled individuals the transfer of spermatozoa is theoretically more likely, but in our experience permanent pairing appears to be the exception rather than the rule. As a matter of fact, all females (several hundred) which we examined, contained spermatozoa in their seminal receptacles, although relatively few were mated at the time. Spermatozoa pass down the uterus and are stored in the seminal receptacle. On discharge from the

TEXTFIGURE III



TEXTFIGURE 3.—Lateral view of mature female *S. mansoni*, showing numerous agglomerations of unorganized egg material in the distal end of the uterus. Camera lucida of living worm, $\times 80$.

GRABADO DEL TEXTO No. 3.—Hembra adulta mansónica, enfocada lateralmente, donde se observan numerosos acúmulos de sustancia ovular sin organizar en el extremo distal del útero. Dibujo en cámara clara de un verme vivo, $\times 80$.

ovary into the proximal end of the oviduct the egg cell is fertilized. It is then carried up to the oötype, where yolk material and a shell are layed on. The egg is then discharged into the proximal end of the uterus, where it partially matures. It seems likely that the egg is ordinarily passed through the long muscular uterine tube and discharged only when the female elongates into a small venule, so that constriction of the anterior portion of her body sets in motion a progressive rythmic constriction of the uterine wall, thus forcing the egg outward.

In view of the statement of Leiper¹, that the mature females contain only a single egg *in utero*, and of Brumpt²¹ (p. 283), that the females have normally only a single egg *in utero* and very rarely two, and the contradictory observations of Fairley³ (p. 294), who stated that he repeatedly saw three eggs in the uterus of the same female, and six eggs deposited in one small venule, "presumably all deposited in succession by one female worm", it was desirable to make observations on this point. For this study we utilized the living worms from the mesenteric venules of the monkey sacrificed on the 55th day. Some were examined while still within the blood vessels; others were examined immediately after perfusing the worms out of the venules. Of 364 worms individually studied, all but 18 contained eggs. Of the total number only 9 were mated with males, and all of these coupled females contained eggs. The great majority contained only a single egg each. The few exceptions noted contained the following egg masses; one with 25 well-formed eggs; one with 17 well-formed eggs; one with 15; two with 12 each; one with 8; one with 6; one with 5; one with 4; one with 3, and fourteen with conglomerate masses of egg material (text fig. 3), more or less stuck together like the ~~fertile or infertile agglomerates~~ of *Ascaris lumbricoides* eggs occasionally seen in human feces. It will be noted, therefore, that only 6.6 per cent of the females contained more than one egg each and that the majority of these were abnormally formed egg masses. Similar observations on another monkey infection and two rabbit infections confirmed our first findings, except that in more chronic infections multiple egg masses and agglomerates were rarer. We may conclude, therefore, that the normal female *S. mansoni* usually has, at any one time, only a single egg *in utero*.