With few exceptions, parasites of the genus Eimeria develop in the epithelial cells of the intestine. In the case of E. stiedae of rabbits and E. truncata of geese, the oocysts excyst in the digestive tract but the parasite develops in epithelial cells of the bile duct and kidney tubules respectively. Long (1970b) reported that E. tenella could develop to the second generation schizont stage in the liver of chickens treated with dexamethasone. The development of Eimerian sporozoites in organs other than the intestine after excystation in the digestive tract may indicate that these sporozoites are carried out from the intestinal mucosa to these organs. A mechanism similar to that obtained here after the intraperitoneal injection of proteose peptone and other irritants may operate in these situations.

The transport of sporozoites of E. necatrix in intraepithelial lymphocytes may have some implication for the control of the disease. Oral immunization of chickens using antigenic parasite products may be of particular importance in interfering with the parasite development and consequently prevention of the disease.

location and the direction of movement of these cells may influence the outcome of an infection.

The i.p. injection of an irritant results in massive infiltration of leucocytes into the peritoneal cavity (Figs. 3-30, 3-31, 3-32). It has also been demonstrated that an i.p. injection of antigen stimulates the production of specific antibody-containing cells and a protective local immunity in the gut (Husband, 1978; 1980). These findings suggest the existence of a relation between the intestinal mucosa and the peritoneal cavity. Therefore the i.p. injection of proteose peptone and other irritants may attract the infected intraepithelial lymphocytes to the peritoneal cavity of chickens and delay their arrival at crypt epithelial cells. This would delay the development of the subsequent stages of the life cycle.

According to Rose (1973), parenteral inoculation of Eimerian sporozoites in many different sites often results in an intestinal infection. Fitzgerald (1962) successfully infected calves with oocysts of E. bovis by injecting them i.p. but failed to infect calves by injecting oocysts subcutaneously, i.m. or i.v. Similar experiments have been conducted with chicken coccidia. The oocysts of E. maxima, E. acervulina, E. necatrix and E. tenella were administered in large numbers intramuscularly, i.p. and i.v. Only chickens inoculated i.p. passed oocysts in their feces but the prepatent period was prolonged (Davis and Joyner, 1962; Sharma and Reid, 1962). If these experiments were properly conducted and extraneous infection did not actually occur, then it could be concluded that excysted sporozoites can reach the intestinal mucosa from the peritoneal cavity.

acervulina in macrophage-like cells in villar lamina propria during the early hours post infection. In the present study results obtained using the special histological stain for sporozoites in intestinal sections 3 to 24 hours after infection were in agreement with the results obtained by Van Doorninck and Becker (1957) who showed that sporozoites of E. necatrix were transported from villous epithelial cells to the crypts area by macrophage-like cells. Accordingly, the possibility that macrophages with sporozoites in them may infiltrate the peritoneal cavity as a result of irritant injection was investigated. Since silica particles are known to kill macrophages specifically (Pearsall and Weiser, 1968; Allison, 1978) it was used in conjunction with proteose peptone injection to study its effect on parasite development. Results of this experiment showed that the i.p. injection of silica did not modify the effect of the i.p. injection of proteose peptone or stop parasite development. This experiment suggested that sporozoites were not transported by macrophages but by some other kind of cells. Electron microscopic examination of the same tissues confirmed that those cells were intraepithelial lymphocytes and not macrophages as suggested by Van Doorninck and Becker (1957).

Chickens inoculated with equal numbers of E. necatrix oocysts may not show the same lesion severity. In addition, histological examination of infected intestinal tissues taken at 5 days post infection may show mature and immature second generation schizonts and, in some instances even first generation schizonts. These findings indicate that parasite development is not synchronous. Since sporozoites may depend on intraepithelial lymphocytes for their transport from villar to crypt epithelial cells, the availability,

Heterophils were therefore, not depleted as a result of the i.p. injection of proteose peptone. Thus, the association between heterophil infiltration and lesion severity was not resolved by these experiments.

Experiments using i.p. injection of proteose peptone and other irritants were continued in order to understand the effect of peritonitis on the development of the parasite. Peak oocyst production in proteose peptone injected chickens was at least two days behind that of the control chickens. In addition, they produced fewer oocysts in chickens inoculated with 1×10^3 oocysts than the control group, though this decrease was not statistically significant. This reduction in oocyst production was not significant probably because of the great variation in the number of oocysts produced by individual chickens in both groups. These results only confirmed the earlier finding that the life cycle of E. necatrix was delayed by an i.p. injection at the time of inoculation but did not give us a definite answer as to whether some parasites are killed before oocyst production. Other irritants like starch, Sephadex and thioglycolate broth when used to induce peritonitis had the same effect on the parasite as proteose peptone indicating that the key factor in parasite retardation was the induction of peritonitis rather than proteose peptone itself.

It was also found that the delay was in the development of the first generation schizonts rather than of the second generation schizonts. It was therefore believed that the sporozoites and their transport from villous epithelial cells to the crypts was affected by the i.p. injection of various irritants. Many workers (Pattillo, 1959; Doran, 1966; Challey and Burns, 1959; Van Doorninck and Becker, 1957) have reported seeing the sporozoites of E. tenella, E. necatrix and E.

DISCUSSION

Intraperitoneal injections of proteose peptone were used initially to isolate large numbers of heterophils (see Chapter 2) from peritoneal exudate in order to produce anti-heterophil serum in rabbits. The failure of anti-heterophil serum to reduce the number of circulating heterophils made it difficult to study their role in lesion severity associated with the development of second generation schizonts of E. necatrix. Since very large numbers of heterophils infiltrated into the peritoneal cavity as a result of proteose peptone injection, (see Chapter 2) this method was used to study the effect of an i.p. injection on lesion severity in E. necatrix.

It was found that chickens injected i.p. with proteose peptone had lower mean lesion scores at 5 days post infection than uninjected infected controls. Histological examination of intestinal tissues taken at this time showed that the reduced lesion severity was accompanied by retardation in the development of second generation schizonts. In chickens injected with proteose peptone and sacrificed at 7 days post infection the gross lesions were as severe as those seen in control uninjected chickens given the same dose of oocysts and sacrificed at 5 days post infection. These results suggested that the reduction in severity was not due to the redirection of heterophils away from the gut but due to the slower development of the parasite. This was also indicated in histological sections of bone marrow from proteose peptone injected and uninjected control chickens examined at 5 days post infection. Bone marrow sections of chickens injected with proteose peptone were full with heterophils, while in those of control chickens, the number of heterophils were much lower (Figs. 3-9, 3-10).

TABLE 3-4. Mean Lesion Scores of Chickens Injected with the Various Agents at Different Times Prior to Infection.

Group	Material	Time Route of Injection*	Mean Lesion Score ± S.E.****
1	BCG	zero - i.v.**	5.1 ± 0.74
2	BCG	7 - i.v.	5.9 ± 0.83
3	BCG	14 - i.v.	6.6 ± 0.54
4	*P. acnes*	zero - i.v.	5.2 ± 0.83
5	*P. acnes*	7 - i.v.	5.0 ± 1.0
6	*P. acnes*	14 - i.v.	6.1 ± 0.74
7	*P. acnes*	7 - i.p.***	5.5 ± 0.57
8	*P. acnes*	14 - i.p.	6.0 ± 0.81
9	Proteose peptone	7 - i.p.	5.8 ± 0.83
10	Proteose peptone	14 - i.p.	6.5 ± 0.57
11	Infected controls	uninjected	6.4 ± 0.84

* days before infection
** i.v. = intravenous
*** i.p. = intraperitoneal
Each group had 5 chickens except group 11 which had 10.
**** Standard error

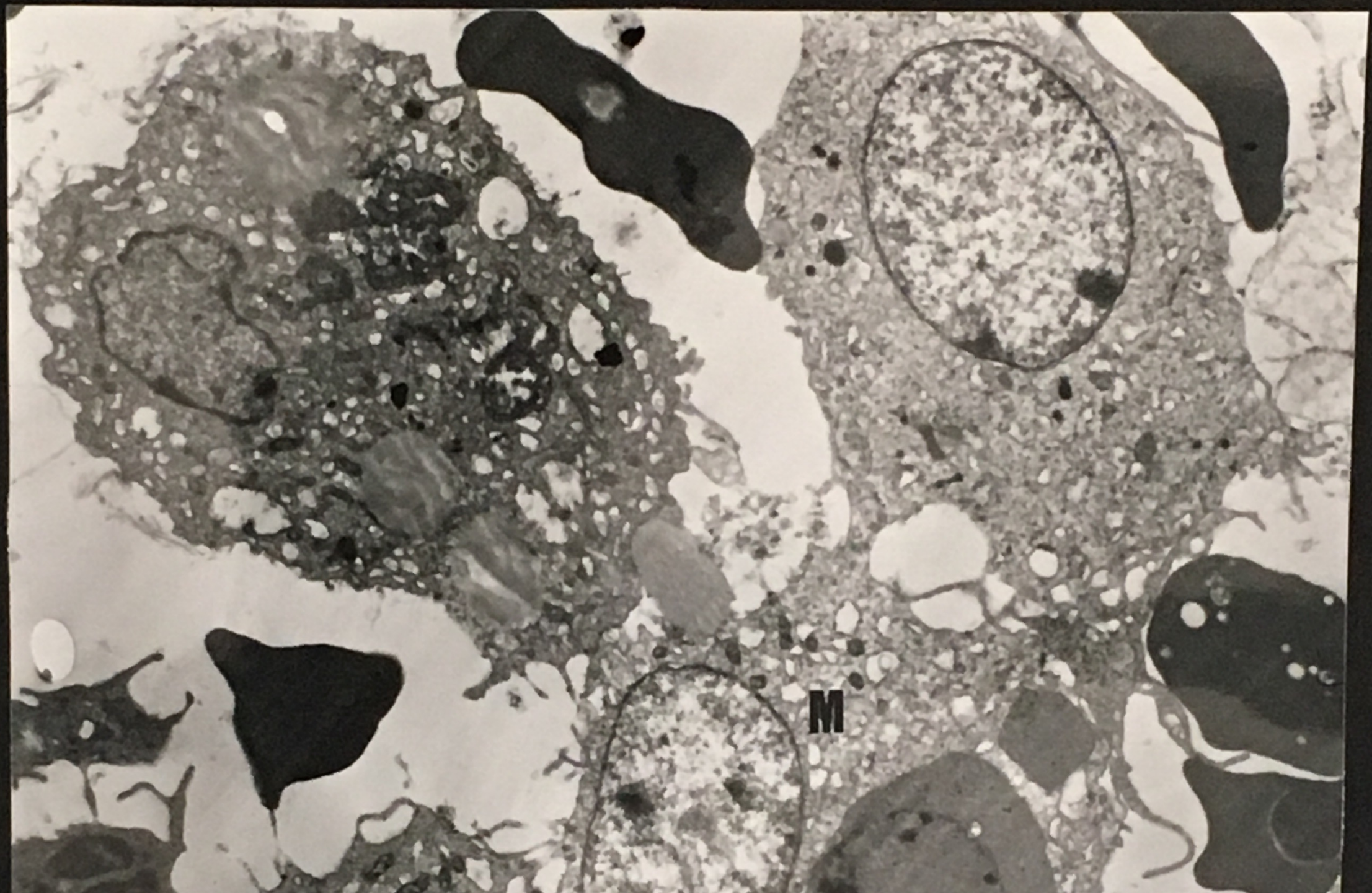
M

Figure 3-32. Electron micrograph of pelleted peritoneal exudate cells taken 48 hours after intraperitoneal injection of proteose peptone. At this time the peritoneal exudate contains mainly mononuclear cells (M). (x7800)

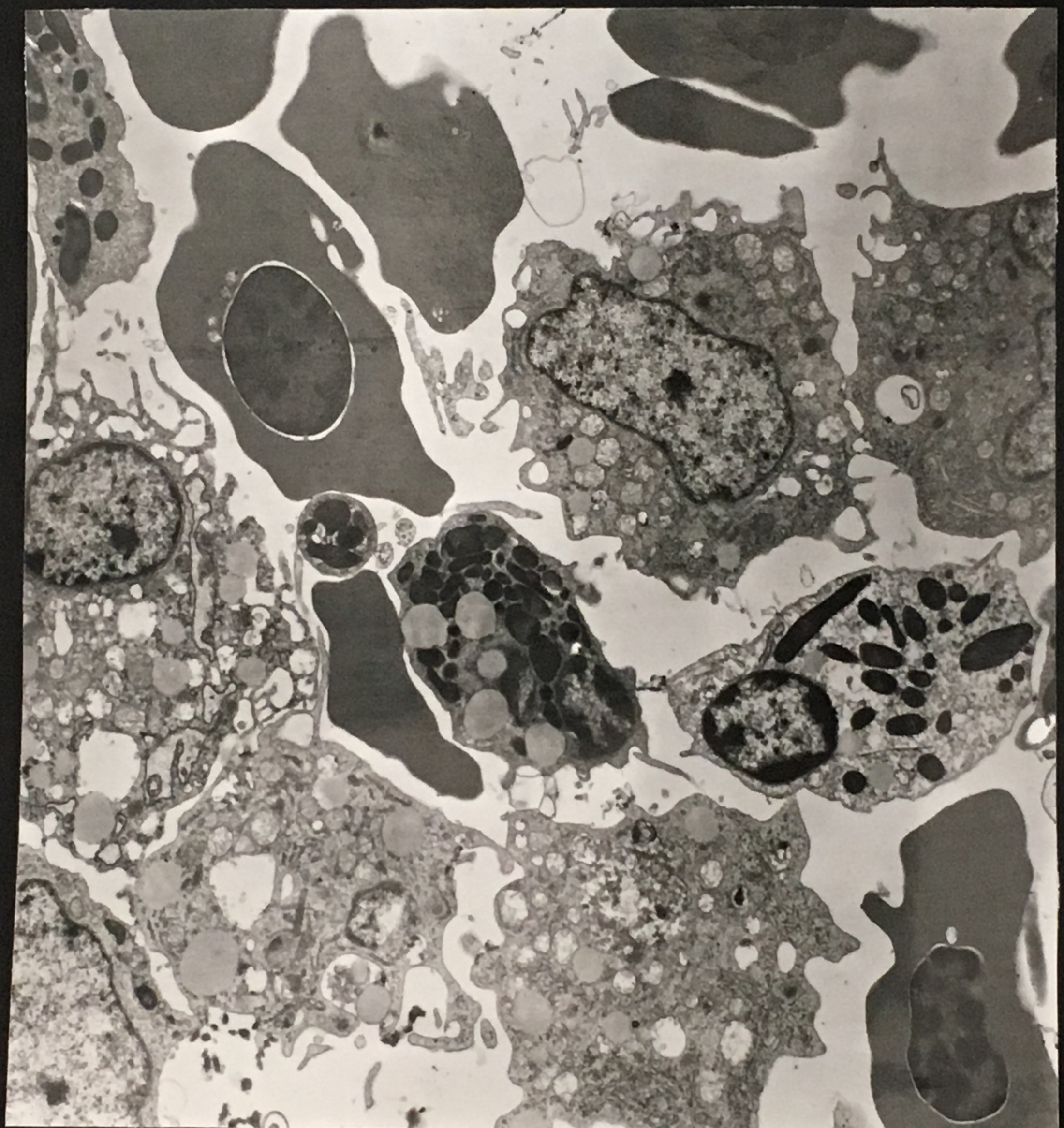

Figure 3-31. Electron micrograph of pelleted peritoneal exudate cells taken 24 hours after intraperitoneal injection of proteose peptone. Note the reduction of heterophil numbers as compared to Figure 3-30. (x7800)

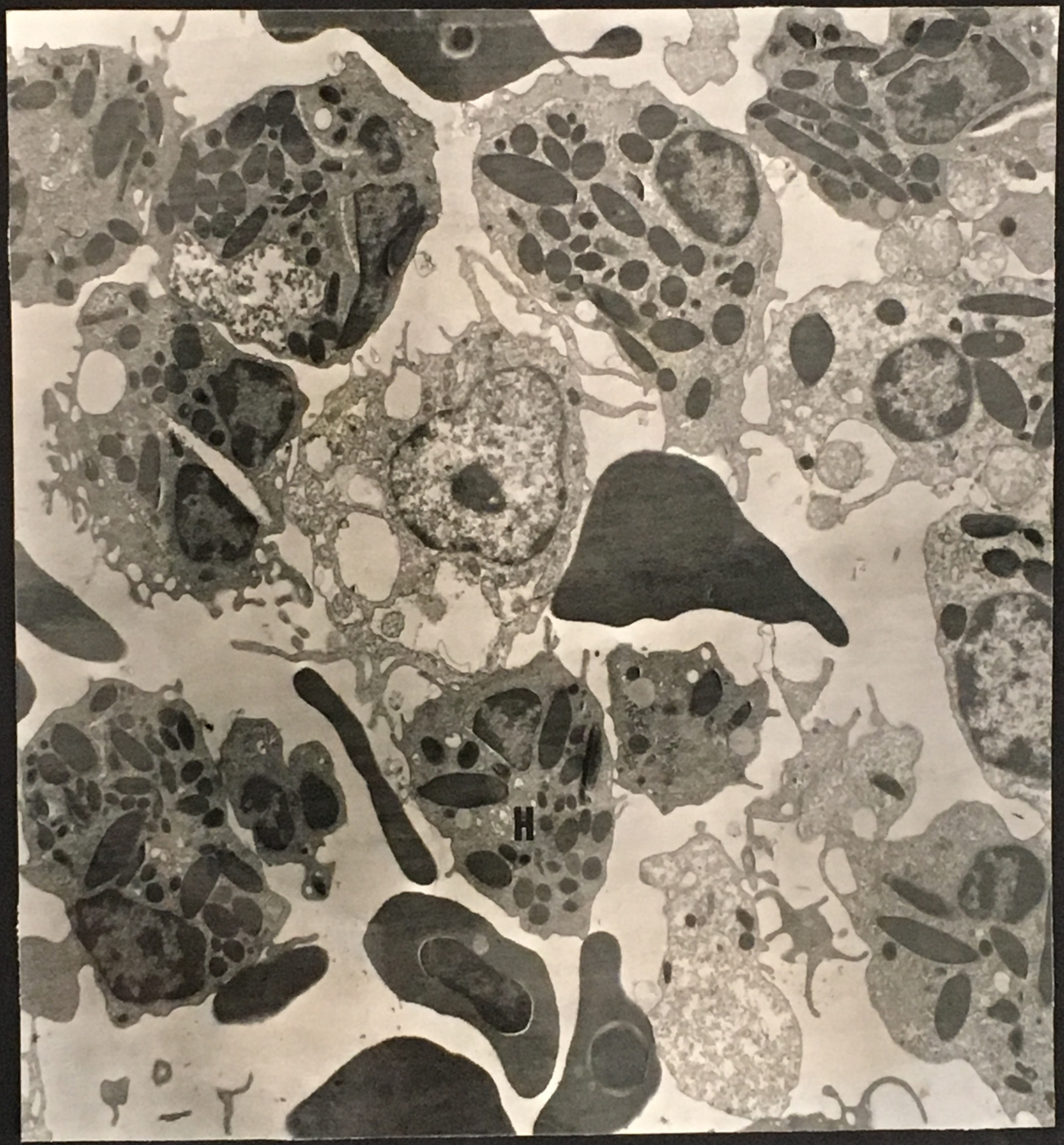
H
٢-٢٥

Figure 3-30. Electron micrograph of pelleted peritoneal exudate cells taken 12 hours after intraperitoneal injection of proteose peptone. Note the high proportion of heterophils (H) at this time (x 7800)