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Effects of Age on Growth and Food Efficiency Response in Rats Infected with Tapeworm Larvae[1] (38175)

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Several groups of investigators (1–3) have shown that the growth rates of young rapidly growing rats cannot be increased by infection with plerocercoids of the tapeworm *Spirometra mansonoides*. However, Steelman *et al.* (4) showed that adult female rats respond to plerocercoid infection with a body weight gain similar to that produced by growth hormone (GH). Glitzer and Steelman (2) also observed a marked decrease in pituitary weight (37%) and in plasma GH levels from control values of 27 ng/ml to nondetectable levels at 10 days following worm infection. These observations have been confirmed by Garland and Daughaday (5).

Steelman and Morgan (6) infected 22 day old castrated male rats with plerocercoids and observed a body weight increase similar to that obtained with GH. No effect was observed on the seminal vesicle and prostate weights but when worm-infected animals were supplemented with methyl testosterone a striking potentiation of the seminal vesicle and prostate weights resulted. These studies point to yet another similarity between pituitary GH and the plerocercoid growth factor (PGF).

In view of these observations, the experiments described in this paper were performed to determine the age at which rats show a growth response to plerocercoid infection. Food efficiency measurements were also made and the skeletal muscles of the host rats were analyzed for moisture and lipid content. If plerocercoid infection should produce appreciable increases in the body weight and food efficiency of older rats without undesirable effects on skeletal muscle, similar studies would be indicated for selected commercial animals.

Methods. Thirty-five day-old Holtzman male rats weighing approximately 100 g were castrated at the Hormone Assay Laboratories (Chicago) and held for 1 wk prior to shipment. Intact male rats of the same age, strain, and approximate weight were included in the same shipment. Eighty day-old female rats of the Holtzman strain and weighing approximately 200 g were also used. Upon arrival, all animals were housed in a temperature-controlled animal room with a 12 hr light/12 hr dark cycle and were fed a purified diet containing 30% casein, 10% corn oil, 4% salts, 56% sucrose and ample amounts of vitamins (7). The animals were killed by decapitation, blood was collected and serum samples were pooled for total lipid (8) and triglyceride (9) measurements. The gastrocnemius muscle from the right rear leg was carefully dissected out and analyzed for moisture and total lipid content.

All stages of the life cycle of *S. mansonoides* were maintained in our laboratory according to the techniques of Mueller (10).

Porcine growth hormone (1 IU/mg) was purchased from Calbiochem.

Results. Intact female rats. In Experiment 1 intact female rats were fed the purified diet for 1 wk prior to the start of the experiment. At 96 days of age, 7 rats were infected sc with 10 plerocercoids each and 8 animals began to receive daily sc injections of 0.5 IU of porcine GH/rat. Eight additional females remained untreated

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TABLE I. Plerocercoid and GH Induced Responses in Adult Intact Female and Castrated Male Rats.

Expt. No.	Group	No. Rats	Initial age (days)	Days on experiment	Average body weight (g)				Food efficiency g wt gain g food	Serum[a]	
					Initial	Final	Increase ±S.E.M.			TG (mg/100 ml)	Lipids (mg/100 ml)
1	Female controls	8	96	18	219	241	22 ± 2.4		0.11	97	340
	Females + plerocercoids[b]	7	96	18	218	255	37 ± 2.9	$P < .005$[d]	0.15	193	490
	Females + GH[c]	8	96	18	220	243	23 ± 2.7	N.S.	0.11	107	370
2	Female controls	10	114	19	241	250	9 ± 3.1		0.05	194	460
	Females + worms[b]	10	114	19	236	269	34 ± 1.3	$P < .005$	0.13	390	730
	Females + GH[c]	10	114	19	238	250	12 ± 2.0	N.S.	0.06	143	440
3	Castrated male controls	10	84	24	300	352	52 ± 4.5		0.14	157	478
	Castrated males + worms[b]	10	84	24	282	345	63 ± 4.8	$P < .05$	0.15	170	433
	Castrated males + GH[c]	10	84	24	301	345	44 ± 6.4	N.S.	0.12	123	435
	Intact males	10	84	24	341	399	58 ± 3.8	N.S.	0.15	147	468
4	Castrated male controls	5	108	27	341	381	40 ± 4.5		0.10	150	423
	Castrated males + worms[b]	5	108	27	345	440	95 ± 4.5	$P < .005$	0.17	285	630
	Intact males	4	108	27	384	430	46 ± 4.9	N.S.	0.10	—	—

[a] Although individual serum samples were collected, equal volumes from individual rats were pooled (1 ml/sample) for analysis.

[b] 10 plerocercoids/rat injected sc.

[c] 0.5 IU porcine GH/rat/day injected sc.

[d] The significance of body weight increase over the respective control group was calculated in each experiment using Student's t test.

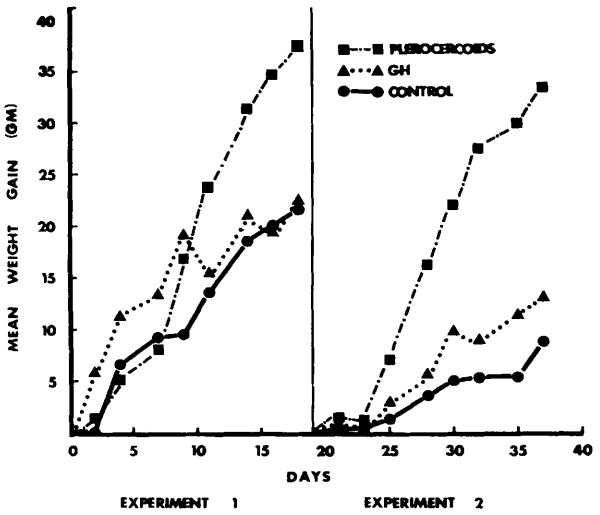

FIG. 1. Body weight gains in female rats either infected with 10 plerocercoids/rat, injected sc with 0.5 IU GH/rat/day or injected with physiological saline (controls). Rats in Experiment 1 were 96 days old at the beginning of the experiment and were sacrificed 18 days later. Fresh groups of animals were started on day 18 (age 114 days) and were sacrificed 19 days later on day 37 (Experiment 2).

and served as controls. After 18 days the animals were sacrificed and the data collected are summarized in Table I and Fig. 1. Worm-infected animals showed a greater body weight gain (37 g) than controls (22 g) but GH produced no significant change (23 g). In Experiment 2, three groups of 114 day-old females were started on the same regimen and were sacrificed 19 days later. The controls in this age group grew only 9 g whereas worm-infected animals grew an average of 34 g in the same 19 day time period (Table I and Fig. 1). Once again, GH produced no significant effect on growth. Both age groups of infected animals also utilized their food more efficiently for growth than either control or GH-treated animals. There were no significant differences in the total lipid and moisture content of the gastrocnemius muscle of worm-infected and control animals, although worm-infected animals had considerably higher serum total lipid and triglyceride concentrations (Table I).

Male rats. In Experiment 3, castrate and intact male rats of the same age (77 days) were held for 1 wk on the purified diet prior to the start of the experiment. Ten castrate rats (84 days old) were then injected sc with 10 pleroceroids each and 10 rats began to receive daily injections of 0.5 IU of porcine GH/rat. Ten intact and 10 castrated rats remained untreated and served as controls. After 24 days the animals were killed. The data are summarized in Table I. Worm-infected castrated males showed a small but significant ($P < .05$) increase in body weight (63 g) over castrate controls (52 g) but GH produced no effect (44 g gain). Three new groups of 108 day-old animals were started (Experiment 4) and sacrificed 27 days later. Although control castrate and intact animals continued to grow (40 and 46 g, respectively), the worm-infected animals grew substantially more (95 g) and utilized their food more efficiently (Table I and Fig. 2). Serum total lipid and triglyceride concentrations were also much higher in worm-infected castrate males (Table I), but as in the case of the female rats, there were no differences between groups in the lipid and moisture content of the gastrocnemius muscle.

Discussion. The data obtained in these experiments show that slowly-growing intact female rats approximately 96–133 days old can be made to grow faster than unin-

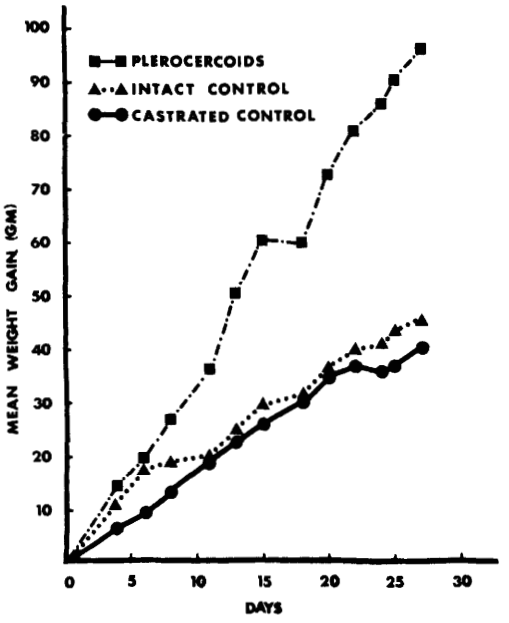

FIG. 2. Comparison of body weight gains in castrate male rats infected with 10 plerocercoids/rat with control castrate and intact male rats (Experiment 4). Rats were 108 days old at the beginning of the experiment and were sacrificed 27 days later.

fected controls and that they utilize their food more efficiently for growth. Although plerocercoid infection had little effect on fairly rapidly growing castrate male rats (84 days old), growth stimulation and increased food efficiency were quite marked in older (108 days old) infected males whose growth rate had slowed. Preliminary studies with slowly growing 104–127 day old intact male rats indicate that growth can also be stimulated by plerocercoid infection (unpublished data).

Growth hormone has been shown to mobilize fat depots both *in vivo* and *in vitro* (11) and Azizi *et al.* (12) have found that plasma triglycerides become elevated in normal young men and older diabetic men and women after human GH administration. The preparation of porcine growth hormone used in our experiments produced no significant change in the body weights of either female or castrate male rats and it had no effect on the serum total lipid and triglyceride concentrations of these animals. On the other hand, plerocercoid infection produced a large increase in both serum triglyceride and total lipid concentrations of the female and castrate host animals. Although there is no immediate explanation for the worm effect, it is interesting to note that Meyer *et al.* (13) found that the plerocercoid of *S. mansonoides* lacks the mechanism for *de novo* synthesis of its sterols and long chain saturated and unsaturated fatty acids. On the other hand, the plerocercoid can synthesize its own triglycerides, sterolesters and phospholipids with the use of exogenous sterols and fatty acids. Since the fatty acid composition of the parasite closely resembles that of its host (13) it is interesting to speculate that the worm may alter the lipid metabolism of its host to provide the substrates it needs for its own metabolism.

The rat growth and food efficiency responses to plerocercoid infection and the lack of any effect on the moisture and lipid content of skeletal muscle suggest the need for comparable studies in selected commercial animals. It would be interesting to know whether body weight gain and food efficiency can be improved in feedlot animals by PGF administration and whether PGF effects the quality and composition of the meat (skeletal muscle). Since the plerocercoid growth factor (PGF) also has prolactin-like activity (14), studies should be made of the effects of PGF on milk production, egg production, reproduction, litter size and the birth weight and growth rate of offspring.

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1. Mueller, J. F., and Reed, P. J., J. Parasitol. 54, 51 (1968).

2. Glitzer, M. S., and Steelman, S. L., Proc. Soc. Exp. Biol. Med. 138, 610 (1971).

3. Ruegamer, W. R., and Mueller, J. F., Proc. Soc. Exp. Biol. Med. 143, 133 (1973).

4. Steelman, S. L., Glitzer, M. S., Ostlind, D. A., and Mueller, J. F., Recent Progr. Hormone Res. 27, 97 (1971).

5. Garland, J. R., and Daughaday, W. H., Proc. Soc. Exp. Biol. Med. 139, 497 (1972).

6. Steelman, S. L., and Morgan, E. R., Endocrinology **90**, 315 (1972).

7. Ruegamer, W. R., and Mueller, J. F., J. Nutr. **103**, 1496 (1973).

8. Ratliff, C. R., Culp, T. W., and Gevedon, R. E., "Advances in Automated Analysis" (L. R. Skeggs, Jr., ed.), Vol. 1, p. 117. Media Inc. (1970).

9. Kessler, G., and Lederer, H., "Automation in Analytical Chemistry" (L. R. Skeggs, Jr., ed.), p. 341. Media Inc. (1966).

10. Mueller, J. F., J. Parasitol. **52**, 437 (1966).

11. Manchester, K. L., "Growth and Growth Hormone" (A. Pecile and E. E. Mueller, eds.), p. 150. Excerpta Medica (1972).

12. Azizi, F., Castelli, W. P., Raben, M. S., and Mitchell, M. L., Proc. Soc. Exp. Biol. Med. **143**, 1187 (1973).

13. Meyer F., Kimura, S., and Mueller, J. F., J. Biol. Chem. **241**, 4224 (1966).

14. Phares, C. K., and Ruegamer, W. R., Proc. Soc. Exp. Biol. Med. **143**, 147 (1973).

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