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Migration of lymphoblasts to the small intestine

I. EFFECT OF *TRICHINELLA SPIRALIS* INFECTION ON THE MIGRATION OF MESENTERIC LYMPHOBLASTS AND MESENTERIC T LYMPHOBLASTS IN SYNGENEIC MICE

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Summary. The migration of [^{125}I]UdR-labelled mesenteric lymph node cells in NIH strain mice at various times after infection with the intestinal parasite *Trichinella spiralis* was investigated. *T. spiralis* produced an enhanced accumulation of mesenteric immunoblasts in the small intestine at 2 and 4 days after infection but not at later times. The enhanced migration occurred when using cells from both uninfected and infected donors, denoting an absence of antigenic specificity. This effect is not secondary to a reduced arrival of cells at sites away from the gut in infected mice, but to a primary increase of the arrival in the small intestine. Mesenteric T lymphoblasts (separated on a nylon-wool column) migrated to the small intestine of uninfected recipients and appear to be a major portion of the population which migrate to the gut of infected recipients. Our results were confirmed using ^{51}Cr to label mesenteric cells. We conclude that the parasite causes the small intestine to become more attractive or retentive for mesenteric blast cells early during infection.

INTRODUCTION

It is known that some lymphoblasts do not recirculate *in vivo* but extravasate from the circulation and lodge in non-lymphoid tissue. Lymphoblasts from the thoracic duct (Gowans & Knight, 1964; Delorme, Hodgett, Hall & Alexander, 1969; Halstead & Hall, 1972; Hall, Parry & Smith, 1972) and mesenteric lymph nodes (Griscelli, Vassalli & McCluskey, 1969; Guy-Grand, Griscelli & Vassalli, 1974; Parrott & Ferguson, 1974; McWilliams, Phillips-Quagliata & Lamm, 1975; Parrott, Rose, Sless, de Freitas & Bruce, 1976) have been shown to migrate to the lamina propria of the small intestine. Various factors regarding this phenomenon are poorly understood, e.g. the relative contribution made by T and B lymphoblasts and the role of antigen in specifically recruiting primed cells. Strong evidence suggests B lymphoblasts migrate to the lamina propria (Halstead & Hall, 1972; Guy-Grand *et al.*, 1974). However, the extent of migration of T lymphoblasts to the small intestine remains controversial (Parrott & Ferguson, 1974; Guy-Grand *et al.*, 1974).

If migration of lymphoblasts to the intestine has any immunological significance it might be expected to be reflected in the mechanisms that allow expulsion and consequent immunity to intestinal parasitic infections. It has been found that immunity to *Nippostrongylus brasiliensis* (Keller & Keist, 1972;

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Kelly & Dineen, 1972; Dineen, Ogilvie & Kelly, 1973) and recently *T. spiralis* (Love, Ogilvie & McLaren, 1976) in the rat, can be transferred using mesenteric lymph node cells. Here we investigate the migration of [^{125}I]UdR-labelled mesenteric lymphoblasts and mesenteric T lymphoblasts in mice, at various times after infection with *T. spiralis*.

MATERIALS AND METHODS

Mice

Eight to 14-week-old mice of the inbred strain NIH (Carworth Europe Laboratory Animals, Alconbury, Huntingdon) of both sexes were used. Mice of the same sex and age were selected for each experiment.

Infections with T. spiralis

Infective larvae were recovered from muscle by artificial pepsin/HCl digest and approximately 450 viable larvae administered to each mouse by oral intubation.

Cell suspensions

Mesenteric lymph node cells (MLN) were gently dispersed by teasing with forceps and scalpel in Eagle's minimum essential medium (Eagle's MEM) and filtering the resultant suspensions through stainless steel mesh. The number of viable cells was assessed by their ability to exclude 0·2 per cent Eosin. All suspensions were prepared at 0–4°.

Preparation of T lymphocytes

A nylon-wool column was used to separate T cells according to the method of Greaves & Brown (1974). The number of T cells in the eluate was estimated by performing a cytotoxic assay with rabbit-anti-mouse thymocyte sera.

Preparation of rabbit anti-mouse thymocyte serum (ATS)

New Zealand White rabbits were given three intravenous injections consisting of 3×10^8, 4×10^8 and 6×10^8 CBA thymocytes, at intervals of 2 weeks and 1 week and bled 1 week after the last injection. The serum was complement deactivated by heating at 56° for 20 min. It was absorbed twice with erythrocytes and liver cells from CBA mice and twice with erythrocytes and liver cells from NIH mice (at a volume of cells to serum of 1:3).

Testing of the anti-thymocyte serum and cytotoxicassay

The anti-serum was tested at various dilutions against bone-marrow cells, inguinal lymph node cells and thymocytes from NIH mice. 0·1 ml of anti-serum was added to 10^6 nucleated cells and incubated at 37°. Control cell suspensions were incubated in normal (absorbed) rabbit serum. After 30 min, 0·1 ml of guinea-pig complement (1/10 guinea-pig serum, Wellcome Laboratories) was added to the cells and serum and they were incubated for a further 45 min at 37°. The cells were washed, resuspended in medium and their viability assessed by exclusion of 0·2 per cent eosin.

The number killed specifically by ATS was calculated as follows:

$$\text{Percentage killed} = \frac{\text{percentage dead (ATS)} - \text{percentage dead (control)}}{100 - \text{percentage dead (control)}} \times 100.$$

The results, using a 1/20 dilution of ATS were thymocytes 98% bone-marrow cells 4·8% and peripheral lymph node cells 63%. The anti-serum was used routinely at 1/20 dilution.

In vitro *labelling of lymphocytes*

In order to label cells undergoing DNA synthesis, mesenteric lymph-node cells were incubated with the analogue of thymidine, [^{125}I]5-iodo-2′deoxyuridine ([^{125}I]UdR, 100 μCi/μg) obtained from the Radiochemical Centre, Amersham. They were incubated in Eagle's MEM containing 0·5 μCi/10^7 cells/ml for 60 min at 37° in a gently shaking water bath. In order to label small lymphocytes as well as blast cells, mesenteric lymph-node cells were incubated with radioactive chromium (sodium chromate, ^{51}Cr) (Radiochemical Centre, Amersham) at a concentration of 50 μCi/10^8 cells/ml of MEM for 30 min at 37° in a gently shaking water bath. After labelling, cells were washed three or four times in medium, and the number of viable cells estimated by using 0·2 per cent Eosin. Cell suspensions were adjusted to a concentration of $5–6 \times 10^7$ viable cells/ml, and a total of 2×10^7–$2{\cdot}5 \times 10^7$ cells were injected into the lateral tail vein. Only cell suspensions with a viability greater than 80 per cent were injected. An injection dose was retained to count the injected radioactivity. Autoradiography of cell preparations revealed that 6·7 per cent of cells from uninfected donors and 10 per cent of cells from infected (day 4 and day 10) donors had incorporated [^{125}I]UdR.

In vivo *distribution of labelled cells*

At various times (usually 24 h) after cell transfer animals were killed and a variety of lymphoid and non-lymphoid tissues were removed as follows: inguinal and mesenteric lymph nodes, Peyer's patches, small intestine, caecum, large intestine, spleen, liver and lung. The radioactivity in each organ was measured by counting for one minute in a gamma counter (Tracer Lab. Gamma Set 500). The one-minute background counts were subtracted from the organ counts and the amount of radioactivity in an organ expressed either as a percentage of the injected dose or as a percentage of the total radioactivity which was recovered from all the selected organs as follows:

$$\text{percentage of recovered radioactivity} = \frac{\text{c.p.m. in organ}}{\text{total c.p.m. recovered from all tissues}} \times 100.$$

The total amount of activity which was recovered from all the excised organs expressed as a percentage of the injected dose is shown.

Statistical analysis

The significance of differences between experimental and control groups was assayed using Student's *t*-test.

RESULTS

(1) Evaluation of the technique used to measure localisation of blast cells—[^{125}I]UdR

(a) *The low recovery of* [^{125}*I*]*UdR*

In accordance with the results of other workers who have used [^{125}I]UdR in this context (Halstead & Hall, 1972; Hall *et al.*, 1972; McWilliams *et al.*, 1975) it was found that 24 h after the i.v. injection of labelled MLN only 10–20 per cent of the injected dose could be recovered from the major organs. The problem has been discussed by Hall and his colleagues (Halstead & Hall, 1972; Hall *et al.*, 1972) and does not appear to be due to toxicity of [^{125}I]UdR.

(b) *Possible utilisation of free* [^{125}*I*]*UdR by intestinal epithelium*

An additional problem in these experiments is the high rate of intestinal epithelial cell turnover (Clarke, 1973) which is accelerated during nematode infection (Symons, 1965).

We found it impossible to remove all free isotope from cell inocula without such extensive washing as to prejudice cell survival, about 5 per cent of the injected dose remaining in the supernatant. In order to assess the importance of such free isotope as a possible source of error we injected into mice a dose of free [^{125}I]UdR (20,000 c.p.m.) twenty times the normal contaminating dose, but which was equivalent to a typical injection dose of 2×10^7 labelled MLN. Only 1–2 per cent of the injected dose was found in the small intestine. It would seem reasonable, therefore, to conclude that in a normal experiment only one twentieth of 1–2 per cent of the injected radioactivity would be found in the small intestine as a consequence of injecting free isotope. Very similar results were obtained after the injection of labelled dead cells (killed by heating at 56° for 30 min).

(2) Time course of migration of mesenteric lymphoblasts

The localization of [^{125}I]UdR-labelled MLN from mice 4 days after receiving *T. spiralis* injections (day-4 donors) in mice which were infected with *T. spiralis* 4 days previously (day-4 recipients) 1 h, 4h, 12h and 24 h after their intravenous injection is shown in Fig. 1. (Preliminary experiments had shown that day-4 donors produced large numbers of cells and that day-4 recipients produce maximum accumulation of MLN in the small intestine).

The results are expressed both as the percentage injected and percentage recovered radioactivity. Ten

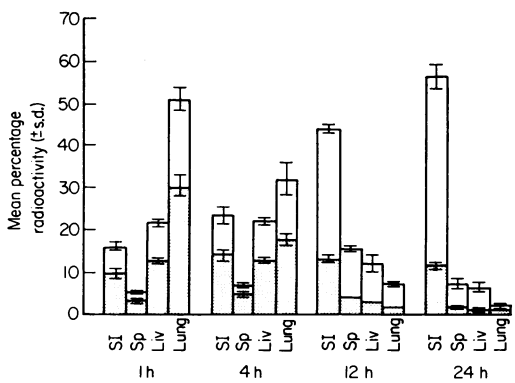

Figure 1. Localization of [^{125}I]UdR MLN in the small intestine (SI) spleen (Sp), liver (Liv) and lung 1 h, 4 h, 12 h and 24 h after their intravenous injection into syngeneic recipients. Results are the mean percent injected radioactivity (stippled area) or recovered radioactivity (open area) from four mice ± s.d.

per cent of the injected radioactivity is recovered from the small intestine (Peyer's patches have been removed) as early as 1 h after cell transfer, and this figure remained relatively constant over the 24-h period. The changes in the distribution of activity between various organs that are observed over 24 h are due to the early transient trapping of cells in the lungs and their subsequent disappearance. After 24 h the counts in the small intestine represented 57 per cent of the total recoverable radioactivity. Since there was little change in the amount of radioactivity that could be recovered from the small intestine over the 24-h period, it was decided to use 24 h routinely as the time period between injection of labelled cells and killing of the recipients.

(3) Migration of mesenteric lymphoblasts in mice infected with *T. spiralis*

T. spiralis lodges in the small intestine and is normally rejected in 9–10 days by adult NIH mice (Bruce, unpublished results). The intestinal localization of [^{125}I]UdR-labelled MLN from day-4 donors in day-4 recipients, day-6 donors in day-6 recipients and day-10 donors in day-10 recipients is shown in Table 1. Compared to the localisation of mesenteric blast cells from normal donors in normal recipients, about twice as much of the injected dose was found in the small intestine of mice 4 days after infection with *T. spiralis* than from normal mice. Thus the presence of *T. spiralis* causes enhanced localisation of mesenteric blast cells in the small intestine, 4 days after infection, but not 6 or 10 days after infection.

In order to ascertain whether this increase represents an increase in the number of cells per milligram tissue or whether it parallels the increase in the weight of the gut, the small intestine from uninfected controls and mice, 4, 6 and 9 days after infection were weighed. The results (Table 2) show that although the gut weighs significantly more 4 days after infection than uninfected gut, the increase is not sufficient to account for the enhanced intestinal localization of MLN blasts at this time. Furthermore, the maximum increase in gut weight occurs 9 days after infection, a time when cell localization has returned to normal.

The enhanced localization of MLN blasts in day-4-infected mice may be due either to the nature of the donor cells and/or the nature of the infected small intestine. To distinguish between these possibilities, the localization of [^{125}I]UdR-labelled MLN from day-4 donors in normal controls, and in mice which were infected 2, 4, 6 and 8 days previously is shown in Fig. 2. Significantly more radioactivity was recovered from the small intestine (less Peyer's patches) of recipients of *T. spiralis* infected 2 and 4 days previously (12·8 per cent and 11·3 per cent of the injected dose respectively) than from the small intestine of uninfected mice (7·2 per cent of the injected dose) or mice which had been infected 6 and 8 days previously.

Thus the effect of *T. spiralis* on the migration of mesenteric lymphoblasts, 2 and 4 days after infection, appears to be mediated via some alteration to the small intestine, making it more attractive or retentive for mesenteric blast cells.

The migration of blast cells to organs other than the small intestine is not significantly altered by *T. spiralis* 4 days after infection (Table 3). The increase found in the mesenteric nodes of infected mice is variable and although not significant in this particular experiment, it has been found to be significant in several other experiments. Similarly the depressed

Table 1. 24-h localization of [^{125}I]UdR-labelled MLN in normal mice and mice infected with *T. spiralis* at various times after infection

Expt	Treatment of mice days after infection		No. of mice	Percent injected dose in small intestine ± s.d.	*P* value*
	Donor	Recipient			
4 Pooled expts	Normal	Normal	19	5·2 ± 2·0	
5 Pooled expts	4	4	25	9·9 ± 2·0	0·00004
2 Pooled expts	6	6	10	5·7 ± 0·8	n.s.
3 Pooled expts	10	10	14	5·1 ± 2·2	n.s.

n.s. = Not significantly different from normal control.
* Compared to normal control.

Table 2. Weights of small intestine at various times after infection with *T. spiralis*

Days after infection	No. of mice	Mean wet weight (g±s.d.)*	Mean dry weight (g±s.d.)†	Wet weight *P* value	Dry weight *P* value
Control	7	0·93±0·09	0·18±0·03		
4	7	1·18±0·18	0·27±0·05	0·006	0·001
6	8	1·28±0·25	0·28±0·06	0·004	0·001
9	8	1·77±0·19	0·38±0·05	0·0000001	0·0000002

* The small intestine was split longitudinally, washed in saline, dried on filter paper.
† After washing as above, the small intestine was dried for 24 h at 100°.

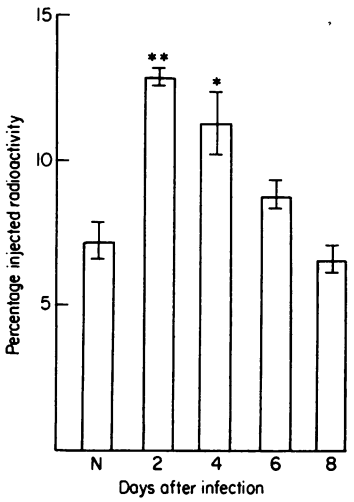

Figure 2. Twenty-four-hour localization of [^{125}I]UdR-labelled MLN from day-4-infected donors in the small intestine of syngeneic uninfected (N) recipients or mice which had received *T. spiralis* infections 2, 4, 6 or 8 days previously. Each point represents the mean percent injected dose from 4 mice±s.e.
* $P<0·05$ or **$P<0·01$ for comparison between uninfected and infected recipients.

localization of cells in the caecum and large intestine of infected mice is not a consistent finding. It is notable that the total amount of recoverable radioactivity is consistently greater in infected (days 2 and 4) than normal mice, and the difference can be accounted for by the greater accumulation of cells in the gut of infected mice.

(4) Specificity of migration of blast cells

It might be expected that during the course of infection, the parasite induces specific proliferation of mesenteric lymph node cells and they or their progeny are preferentially retained in the small intestine of infected mice. This possibility was tested by comparing the ability of cells from normal and day-4-infected mice to migrate to the small intestine of normal and day-4 recipients (Table 4). It can be seen that there was no significant difference in the ability of the two cell populations to migrate to the small intestine of infected mice. Thus there is no evidence that *T. spiralis* specifically causes retention of primed cells in the small intestine at this time.

(5) Migration of ^{51}Cr-labelled MLN in mice infected with *T. spiralis*

We have already eliminated the problem of utilization of free isotope in the gut as a major error in normal mice but it, nevertheless, appeared necessary to check our results at the critical time (day 4) with

Table 3. Localization of [^{125}I]UdR-labelled MLN* in normal mice, and mice infected with *T. spiralis* 4 days previously

Organs	Mean percentage injected dose±s.d.	
	Normal	Infected
MLN	0·8±0·5	1·2±0·4
Peyer's patches	0·5±0·1	0·5±0·1
Small intestine	7·2±1·4	11·2±1·2†
Caecum & LI	2·1±0·2	1·6±0·1
Spleen	1·8±0·5	1·9±0·4
Liver	0·9±0·4	1·0±0·4
Lung	0·4±0·1	0·3±0·1
Total‡	13·7±0·9	17·8±1·5

LI = Large intestine.
* From day-4-infected donors.
† $P<0·05$ compared to normal value.
‡ Total amount of radioactivity that could be recovered from the above organs (plus inguinal nodes) expressed as a percentage of the injected dose.

Table 4. 24 h localization of [^{125}I]UdR-labelled MLN from normal and infected (day-4) donors in normal and infected (day-4) recipients

Treatment of mice		No. of mice	Mean percentage injected dose ± s.d. in small intestine	*P* value*
Donors	Recipients			
Normal	Normal	5	8·4 ± 0·6	
Normal	Infected	4	9·8 ± 0·5	< 0·02
Infected	Normal	4	6·2 ± 0·6	
Infected	Infected	4	8·1 ± 1·0	< 0·02

* Comparison between normal and infected recipients.

Table 5. Twenty-four-hour localization of ^{51}Cr-labelled MLN§ in normal mice, and mice infected with *T. spiralis* 4 days previously

Organs	Mean percentage injected dose ± s.d. Normal*	Infected†
MLN	8·0 ± 1·4	9·7 ± 0·2
Peyer's patches	1·5 ± 0·4	1·6 ± 0·4
Small intestine	1·5 ± 0·1	2·2 ± 0·1‡
Spleen	15·0 ± 0·6	14·6 ± 1·2
Liver	21·0 ± 0·6	21·3 ± 1·2
Lung	2·6 ± 0·2	1·7 ± 0·4
Total	50·2 ± 2·6	50·7 ± 2·4

* Four Mice.
† Five Mice.
‡ $P < 0·05$ compared to normal value.
§ From day-4-infected donors.

another isotope label namely ^{51}Cr. It can be seen that the *in vivo* distribution of ^{51}Cr-labelled MLN (Table 5) is very different from that of [^{125}I]UdR-labelled blast cells (Table 3). Nevertheless, the effect of *T. spiralis* on the distribution of ^{51}Cr-labelled MLN and [^{125}I]UdR-labelled MLN is similar, more radioactivity is recovered from the mesenteric lymph nodes and small intestine of infected mice than normal mice, and there was no significant changes in any other organs.

(6) Migration of separated T lymphoblasts

Previous experiments have used whole mesenteric lymph node suspensions which would be mixtures of T and B cells. Cell inocula from normal and infected donors were found to contain 40–60% T cells using ATS. Mesenteric lymph-node cells from day-4 donors separated on a nylon-wool column were found to be 95% theta positive. When labelled with [^{125}I]UdR they migrated to the small intestine of infected recipients to a similar or even greater extent than unseparated MLN (Table 6, Expt 1). More radioactivity was recovered from infected recipients of T lymphoblasts than from infected recipients of unseparated cells. The increased accumulation of T-MLN in the gut of infected mice was considerably more pronounced than that found using unseparated cells (Expt 2). Also more radioactivity was recovered from infected recipients of T-lymphoblasts (21·0 per cent of the injected dose) than from normal recipients

Table 6. Localization of [^{125}I]UdR-labelled T-lymphoblasts from mesenteric nodes*

	Donor	Recipient	Mean percent injected dose in various organs (s.d.) MLN	Small intestine	Lung	Total recovered
Expt 1	MLN‡	Normal	0·7 (0·4)	5·6 (1·0)	0·57 (0·04)	12·8 (1·4)
	MLN‡	Infected†	0·96 (0·5)	7·4§ (0·83)	0·4 (0·04)	13·8 (1·2)
	T-MLN	Infected†	1·3 (0·4)	9·9 (1·5)	0·24 (0·02)	16·9 (1·7)
Expt 2	T-MLN	Normal	0·6 (0·4)	6·4 (0·7)	0·3 (0·1)	13·0 (1·3)
	T-MLN	Infected†	1·9 (0·6)	12·1¶ (0·5)	1·0 (0·2)	21·0 (1·2)

* Mesenteric blast cells from day-4 donors were separated on a nylon wool column.
† 4 Days after infection with *T. spiralis*.
‡ From day-4 infected donors.
§ $P < 0·05$—Significantly different from normal recipients.
¶ $P < 0·001$—Significantly different from normal recipients.

Table 7. Localization of ^{51}Cr-labelled mesenteric T-lymphoblasts*

Recipient	Mean percent injected dose in various organs (s.d.)					
	MLN	Small intestine	Spleen	Liver	Lung	Total
Normal	13·4 (2·4)	2·1 (0·12)	16·6 (1·2)	10·9 (1·2)	2·1 (0·4)	49·4 (3·0)
Infected†	13·5 (1·0)	2·8‡ (0·3)	13·9 (0·6)	11·5 (1·0)	2·3 (0·4)	48·5 (2·0)

* Mesenteric cells from day-4 donors were separated on a nylon wool column.

† 4 Days after infection with *T. spiralis*.

‡ $P<0·01$, Significantly different from normal recipient.

of the same cell suspension (13·0% of the injected dose). The use of ^{51}Cr to label mesenteric T lymphocytes (separated on a nylon-wool column) produced a different pattern of tissue-distribution (Table 7) to that obtained using [^{125}I]UdR (Table 6). ^{51}Cr-labelled T-MLN preferentially localized in lymphoid tissue and spleen rather than the small intestine. Nevertheless, in confirmation of the results obtained using [^{125}I]UdR, significantly more activity was recovered from the small intestine of infected recipients than from uninfected recipients.

DISCUSSION

The present studies have used the intestinal parasite *T. spiralis* as a tool to investigate the factors that regulate the migration of lymphoblasts to the small intestine. The salient finding of this report is that significantly more mesenteric lymphoblasts accumulate in the small intestine of mice with a day-4 infection of *T. spiralis* than accumulate in the small bowel of uninfected mice or mice with a 6- or 9-day infection. There are two possible explanations: one is that *T. spiralis* produced more 'gut-homing' blast cells in the mesenteric nodes at this time; the second is that it causes the small intestine to become more attractive or retentive for mesenteric immunoblasts. Further experiments demonstrated that cells removed from day-4 donors showed enhanced intestinal migration only in mice 2 and 4 days after infection—migration of these cells to the small intestine of uninfected mice and mice 6 and 8 days after infection was considerably less. Thus we conclude that some feature of the small intestine 2 and 4 days after infection causes it to be particularly retentive of mesenteric immunoblasts.

Infection with *T. spiralis* does increase the weight of the gut but the increase does not coincide with the peak of immunoblast migration. Cells from normal donors migrated to the gut of 4-day-infected recipients as readily as cells from infected donors. This indicates that the migrating cells were not recruited or retarded in the gut as a direct consequence of specific priming to the antigen of *T. spiralis*. This finding of an absence of antigenic specificity has been corroborated recently by Love & Ogilvie (1976) who have also found that intestinal parasites, including *T. spiralis* cause non-specific accumulation of [^{125}I]UdR-labelled rat thoracic duct cells in the small gut. The most likely explanation at present for the accumulation of lymphoblasts produced by *T. spiralis* is a non-specific mechanism, for example, increased blood flow or permeability of the endothelial cells lining the intestinal capillary. Supporting this idea is our preliminary observation (Parrott *et al.* 1976) that orally administered turpentine (an inflammatory agent) also produces an increase in lymphoblast localization in the small intestine. Although inflammation is associated with the pathology of infections with *T. spiralis* (Larsh & Weatherly, 1975) the production of inflammatory lesions and their relation to the course of infection in NIH mice has not yet been established.

We concluded that the increased localization observed on days 2 and 4 after infection is due to an increased retention of blast cells because there was no evidence of redistribution of cells in the tissue of infected as compared with uninfected mice.

The effect of *T. spiralis* or the localization of [^{125}I]UdR-labelled mesenteric cells was also found when using ^{51}Cr. This confirms that the increase on day 4 is not an aberrant result due to utilization of free isotope in the supernatant.

Most previous studies on migration of lymphoblasts to the gut have been concerned with potential IgA forming B blasts (Gowans & Knight, 1964; Halstead & Hall, 1972; Hall *et al.*, 1972; Guy-Grand *et al.*, 1974; Delorme *et al.*, 1969). The present studies have demonstrated that T-lymphoblasts obtained from the mesenteric lymph nodes are also able to migrate to the lamina propria of the small intestine, corroborating the results of Guy-Grand *et al.* (1974). They show moreover that mesenteric T lymphoblasts are more efficient at discriminating between normal and infected gut, than unseparated cells. We have found that *T. spiralis* causes the intestinal retention of mesenteric blasts which would not

normally localize in any tissue, and that this increased retention consists of mesenteric T-lymphoblasts. The theories which have sought to explain the migration of lymphoblasts to the small intestine on the basis of a possible affinity between IgA on the surface of blast cells and cells in the gut which synthesize secretory component (Halstead & Hall 1972; Hall *et al.*, 1972; Guy-Grand *et al.*, 1974) cannot accommodate these observations on T-lymphoblasts. One is still left without a satisfactory explanation to account for the efficiency of blast accumulation in the small intestine since these and many other observations have precluded an influence of positive antigenic attraction.

It is difficult to assess the functional significance of lymphoblast-intestinal migration in the context of nematode expulsions since the nature of the host response to *T. spiralis* is still not certain (Larsh & Weatherly, 1975). Although only circumstantial evidence, the temporal relationship between migration of cells to infected gut on days 2 and 4 and the expulsion of worms on day 9 suggests that the migrating population may be involved with the expulsion process. Expulsion of *T. spiralis* has been found to be T-dependent (Walls, Carter, Leuchars & Davies, 1973) but the nature of the T-dependence is unknown. Recently it has been suggested (Love *et al.*, 1976) that expulsion of *T. spiralis* may involve a two-step operation consisting of antibody damage to worms and subsequent expulsion of damaged worms by sensitized mesenteric lymph-node cells. This process has been shown to result in the expulsion of other nematodes from the small intestine (reviewed in Ogilvie & Love, 1974). It may be, therefore, that T-mesenteric lymphoblasts are involved in the expulsion of antibody damaged *T. spiralis* worms.

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