pipette to a length of precisely 15 mm in the pipette. Each pipette has an aperature just large enough to allow passage of a 21-gauge needle. The pipettes are placed in a test tube rack with their tip suspended into a test tube and incubated at 37 °C. Venous blood was drawn in heparinized tubes from healthy human volunteers. Each volunteer serves as his own control in that each sample of blood was divided into equal portions. To each ml of test sample 0.01 ml of epinephrine (1:1000 dilution) was added. The blood was then incubated for 10 min, mixed by gentle turning and 1 ml aliquots were added to the pipettes and allowed to filter through over 10 min. Each experiment with and without epinephrine was run in triplicate. The number of granulocytes in the blood was measured before and after passage through the column and the percentage of granulocyte adherence was calculated.

Results. The adherence of granulocytes was decreased by 27.9% in the presence of epinephrine (table). This was statistically significant at $p < 0.0025$ level analyzed by the Student t-test.

Discussion. Many clinical conditions with increased susceptibility to infection cannot be explained by decreases in granulocyte phagocytosis or chemotaxis alone and additional granulocyte function defects must be considered. Granulocyte adherence to nylon fibres or glass may be used as measure of their ability to adhere to endothelial walls but this requires further proof.

It has been shown by MacGregor et al.[3] that anti-inflammatory drugs inhibit granulocyte adherence to nylon fibres and suggested that this may be the explanation for their effects in vivo. Other agents have also been shown to inhibit granulocyte adhesion such as ethanol[3], iodoacetamide[5], and EDTA[3,6], cyclic AMP[7], colchicine[4], and glycolitic inhibitors[5], whereas oxidative inhibitors[5] had no effect. The presence of certain ions (magnesium and perhaps calcium)[3,6] appear to be necessary.

Abnormal granulocyte adhesion has been demonstrated in several clinical condititions, i.e. leukemia[8] and acute poststreptococcal glomerulonephritis[9]. In the latter, this abnormality can be followed as a measure of resolution of disease.

The administration of epinephrine in vivo causes the granulocytes to demarginate and enter the circulation. This study showed a decreased granulocyte adhesion to nylon fibres in the presence of epinephrine in vitro suggesting that similar mechanisms may operate in both systems. Granulocyte adhesiveness should be studied in clinical situations associated with an increased susceptibility to infection. Information obtained from these studies may provide better understanding of the role granulocyte adhesion plays in preventing infection.

Effect of Epinephrine on granulocyte adhesion

	Percent adhesion ± SEM
Control	75.7 ± 5.0
Epinephrine	47.8 ± 6.4
	$p < 0.0025$

Each value represents 8 experiments performed on blood from healthy male volunteers.

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Accumulation of 2-μm latex particles in mouse Peyer's patches during chronic latex feeding[1,2]

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Summary. 2-μm latex particles accumulated in macrophages in intestinal Peyer's patches of mice given latex suspensions as drinking fluid for 2 months. The number of particles accumulating was a direct (but nonlinear) function of the number ingested. Some of the latex particles were still present in Peyer's patches 6 weeks after the cessation of latex feeding.

The epithelium of gut-associated lymphoid tissues (GALT; Peyer's patches, appendix, sacculus rotundus) appears to be a route by which small inert particulates pass the mucosal barrier of the mammalian intestinal tract[6,7]. Previous morphological studies have described the migration of carbon, trypan blue, and ferritin particles into the GALT[8–12]. These particulates are small, less than 0.1 μm in diameter, and were seen with the light microscope because of their accumulation into aggregates. The present communication describes the penetration of the mouse Peyer's patch epithelium by much larger particles, namely latex spheres of 2-μm diameter. Latex particles resist degradation in the intestine and are large enough to be seen and counted with the light microscope, permitting a semiquantitative assessment of their uptake and distribution. The results indicate that many thousands of latex particles accumulated in Peyer's patches during chronic feeding of mice with latex suspensions. Furthermore, many of the particles were retained for more than 6 weeks after the cessation of latex feeding.

Materials and methods. 3 water suspensions of polyvinyltoluene latex (mean particle diameter ± SD, 2.02 ± 0.014 μm; particle density, 1.027; identification No. LS-1078-B, The Dow Chemical Co.) containing 1.0, 0.1 and 0.01% solids were given as drinking fluid to 3 groups of 11-week-old female Swiss mice. The mice were given free access to the suspensions and to standard pelleted mouse food. The latex suspensions were given for 61 days followed by a period of 2 to 6 weeks during which the mice received latex-free water. A control group was given tap water to drink. No attempt was made to determine the amount of latex ingested by each mouse; however, the volumes of fluid consumed by the 4 groups did not appear to differ. All mice gained weight normally and appeared healthy.

Uptake and retention of latex in Peyer's patches were demonstrated histologically in preparations of whole Peyer's patches cleared in KOH and glycerol[13] and in methacrylate-embedded sections. In addition latex was recovered from digests of intestinal tissues of latex-fed mice. The entire small intestine was rinsed in saline, separated into Peyer's patch and non-Peyer's patch (remainder) tissue, weighed, minced, and placed in test tubes containing 10 ml 3% KOH. The tubes were agitated until the tissue was completely digested. The material was then centrifuged, washed in water, and the latex recovered by ultracentrifugation of the sediment through a linear density gradient of 0–20% urea. Particles which banded at the same level as known latex were recovered by syringe and an aliquot counted using a Neubauer chamber and an optical microscope. Additional aliquots were deposited on 0.8-μm Nucleopore filters, rinsed with water, and examined at 5000× by scanning electron microscopy. The recovered material contained spheres identical to the stock latex; these were not found elsewhere in the gradient.

Results and discussion. 80 whole cleared Peyer's patches were examined with a wide-field inverted microscope. Latex was unaffected by the clearing process, and individual particles could be seen at different levels in the tissue. Each patch consisted of 2–8 follicles; the dome[14] of each follicle of latex-fed mice characteristically showed an accumulation of latex particles in the central portion. Figure 1 shows such an accumulation in a Peyer's patch from a mouse fed the highest concentration of latex. Particles near the mucosal surface were distributed singly or in small clusters whereas particles close to the serosal surface were mostly in aggregates near the periphery of the follicle. Patches from mice given lower concentrations of latex had fewer particles and smaller aggregates. Cleared patches from control mice contained no particulates resembling 2-μm latex. Latex particles were still visible in diminished numbers in cleared Peyer's patches 6 weeks after termination of latex ingestion. The location of latex in relation to cellular elements could not be determined in cleared preparations, but in plastic embedded sections the particles were seen to be associated with macrophages (figure 2).

The table lists numbers of latex particles recovered after digestion of intestinal tissues and ultracentrifugation of the sediment through a density gradient. Sediments of control tissues centrifuged through the density gradient showed no banding of particles at the latex level. The table shows that Peyer's patch tissue comprised only a small fraction of the weight of the small intestine, but contained the major portion of the latex. The pattern of uptake was that of incomplete saturation. The location of the latex found in the remainder of the intestine has not yet been determined.

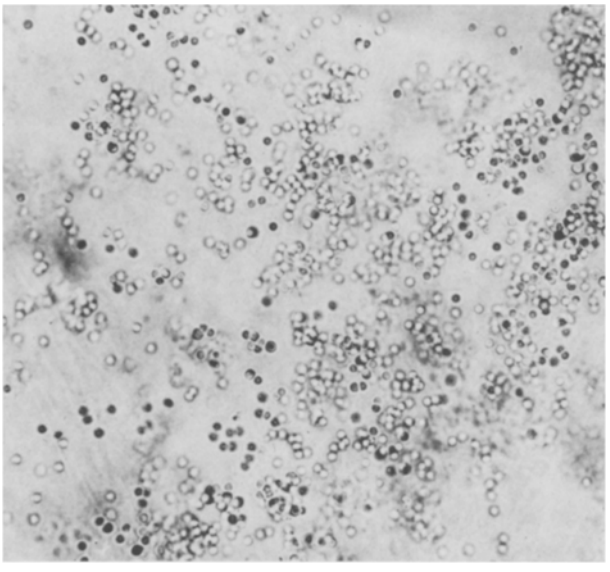

Fig. 1. 2μm latex particles within the dome of a cleared Peyer's patch follicle from a mouse sacrificed 2 weeks after termination of latex feeding. Black circles are latex particles above the plane of focus. Dark shadows are large aggregates of latex below the plane of focus. ×520.

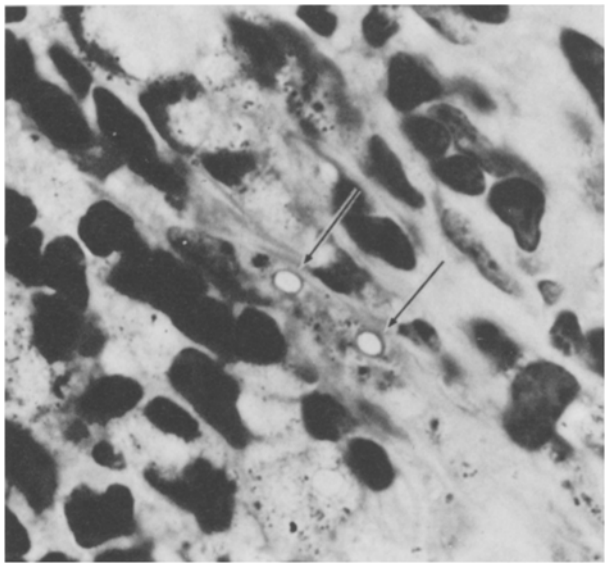

Fig. 2. 2μm latex particles (arrows) within a macrophage process in a mouse Peyer's patch. Distortion of the particles from spherical shape is due to the action of methyl methacrylate which partially dissolves latex during the embedding process. Dark lymphocyte nuclei are visible together with several pale macrophages containing other partially visible latex particles. Section was taken near the serosal surface of a Peyer's patch from a mouse given a suspension of latex as drinking fluid for 2 months. H. & E. ×1500.

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2 The research described in this report involved animals maintained in animal care facilities fully accredited by the American Association for Accreditation of Laboratory Animal Care.

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Quantitative determination of latex recovered from Peyer's patches and remainder of small intestine

No. Latex suspension fed* (% solids)	N	Peyer's patches Tissue weight (g)	No. particles $\times 10^5$	Remainder Tissue weight (g)	No. particles $\times 10^5$
1.0	15	0.054 ± 0.016	2.13 ± 0.66	1.062 ± 0.248	1.78 ± 0.58
0.1	15	0.056 ± 0.013	1.42 ± 0.98	1.078 ± 0.227	1.04 ± 0.29
0.01	15	0.045 ± 0.008	0.18 ± 0.14	1.005 ± 0.176	< 0.05

Values are mean $\pm$ SD; N = number of mice; *Latex suspensions were withheld 2 weeks prior to sacrifice.

These studies indicate that inert particulates of considerable size can cross the mucosal barrier overlying Peyer's patches in intact animals. We estimate that 1 ml of 1.0% latex contained 2.7×10^9 particles and that each mouse given this concentration to drink ingested approximately 7×10^{11} particles in the 61-day test period. Mice receiving lower concentrations of latex ingested proportionately fewer particles. It is clear that the number of particles recovered from intestinal tissue was an extremely small fraction of the number ingested. Nevertheless, when viewed in the perspective of environmental and human health problems, the uptake and retention of even a few particulates by the intestine may be of great importance if the particulates are toxic, mutagenic or carcinogenic.

Rejection of worm load through singly and repeatedly sensitized peritoneal exudate cells during experimental ancylostomiasis

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Summary. Sensitized peritoneal exudate cells from Swiss albino mice donors infected with a single dose of 1000 *A. caninum* larvae could expel a challenge dose of 500 larvae from recipients at a faster rate when compared to cells from repeatedly infected (250 + 250 + 500) donors. However, at 36 h after challenge, the larval expulsion was almost the same in both the groups. Because of the bowel sensitization by the cells, some larvae (not expelled) in the 1st group, readily migrated into muscles where they met allergic immobilization and death due to infiltration of inflammatory cells and their exudates at these sites.

Transfer of immunity (cell mediated immune response or delayed hypersensitivity) from infected donors have been reported for a number of helminthic infections (Larsh et al.[2] with *T. spiralis* in mice, Wagland and Dineen[3] with *Trichostrongylus colubriformis* in isogenic strain of guinea-pigs, Miller[4] with *A. caninum* in experimental pups and Ogilvie and Jones[5] with *N. brasiliensis* in rats using lymphoid cells and Larsh et al.[6,7] with *T. spiralis* in mice and Lang et al.[8] with metacercaria of *F. hepatica* in mice using peritoneal exudate cells). Kim et al.[9] demonstrated that the response can also be produced by antigens of *T. spiralis*. While working on cell-mediated immunity during experimental infection of *Ancylostoma canium*, an attempt was made to investigate the effects of transfer of sensitized peritoneal exudate cells from singly and repeatedly infected donor mice, and the present communication provides evidence regarding development of strong immunity in recipients injected with sensitized cells from the former group of donors.

Material and method. Infective *A. caninum* larvae were cultured following the method of Sen et al.[10] and donor mice were infected according to the following schedule.

Table 1. Experimental schedule of infection to donor mice groups

Day	Sensitizing dose of *A. caninum* larvae to donor groups A (singly infected)	B (repeatedly infected)	C (uninfected control)
0	1000	250	–
7	–	250	–
14	–	500	–
21	Collection and transfer of cells from donors to recipients		
28	Challenge infection of 500 larvae to each recipient		

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