PASSIVE TRANSFER OF ADJUVANT ARTHRITIS BY LYMPH NODE OR SPLEEN CELLS*

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Plates 42 to 48

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Adjuvant arthritis is a unique experimental disorder in which rats develop a migratory polyarthritis and certain other tissue lesions about 10 to 14 days after a single inoculation of standard Freund's water-in-oil adjuvant, or some slight variant of it (1, 2). As investigations of this inducible disorder have proceeded, it has become fairly apparent that the condition is caused by an immunologic response on the part of the inoculated host, although the mechanisms responsible for this are as yet unclear. One chief roadblock, the elimination of an infectious bacterial, mycotic, or mycoplasmal etiology, seemed unlikely when a culturable microbiological agent could not be found with any consistency in either the inflammatory joint tissues or the serum, nor in fact in other tissues or organs (3). Special methods for isolating mycoplasmal or pleuropneumonia-like organisms from the arthritic lesions did not indicate that these agents were involved. Moreover, adjuvant arthritis was readily reproducible in "germ-free" animals (4), although the presence of viruses could not be completely ruled out, even in this experiment.

It was postulated (5) that the disease was a generalized immunologic response, probably of the delayed or cellular type, to one or more antigenic constituents in the tubercle bacillus, since these were essential ingredients of the adjuvant. The sensitizing antigen was probably not tuberculoprotein since in the past 4 years we have used the wax D fraction extracted from the human Canetti strain of tubercle bacilli, instead of whole acid-fast bacilli in the adjuvant mixture, and have had equal or somewhat greater success in inducing arthritic disease with this preparation; *i.e.*, in 85 to 100 per cent or animals in various experiments. Furthermore a lack of parallelism has been noted, in animals inoculated with whole tubercle bacilli in adjuvant, between adjuvant arthritis and cutaneous tuberculin sensitivity (6). Since the wax D fraction, which is chemically classified as a peptidoglycolipid (or a macromolecular glycolipid) (7), usually contains a minimal amount of protein, it is not likely that the latter is the sensitizing agent, although its actual content does vary somewhat

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from one preparation to another. One wax D preparation, especially purified by Dr. Lederer, contained so little protein (if any at all) that it did not induce a tuberculin reaction in guinea pigs injected 3 weeks previously. Yet this preparation very successfully induced adjuvant arthritis.

In earlier experiments, it was not possible to transmit the disease when either sizable amounts of acute phase or convalescent serum (up to 15 ml, intravenously) was transferred from arthritic rats to normal recipients. Furthermore, transfer of the disease process was not previously successful when living sensitized lymph node cells were passed from donors sensitized with adjuvant containing whole acid-fast bacilli to non-inbred recipients even though some of the recipients were previously given whole body irradiation or had been rendered tolerant to specific donor cells (5).

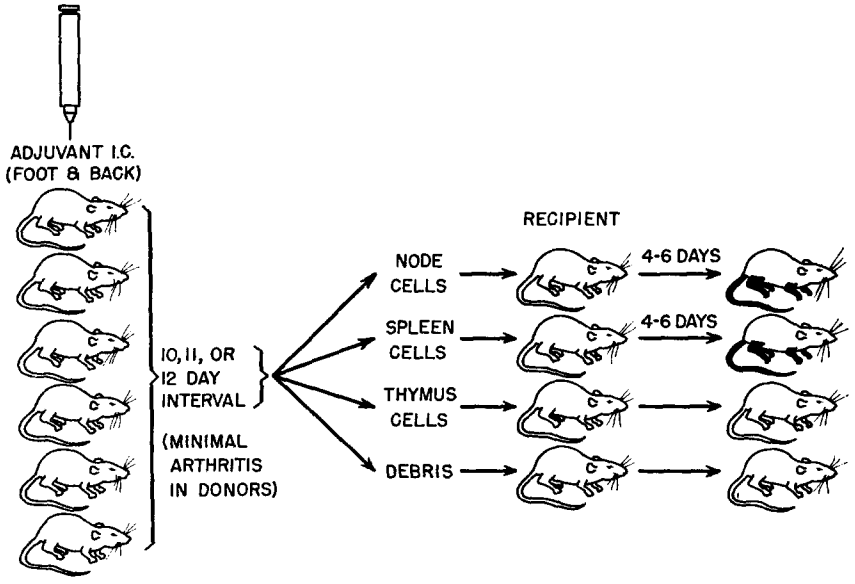
It has now been possible to transfer the arthritic disease passively from sensitized donors to normal recipients under certain conditions, namely (*a*) use of viable lymph node or spleen cells, (*b*) utilization of a highly inbred strain of rats, (*c*) harvesting of cells from donors during certain time intervals after adjuvant injection into them, and (*d*) transfer of sufficient numbers of sensitized viable cells from either the lymph nodes or the spleen, but not from the thymus. These results constitute strong evidence that adjuvant arthritis is an immunological disease which is manifested through a delayed type of reaction, or some variant of it. In this respect, it can be aligned with experimental allergic encephalomyelitis, experimental allergic thyroiditis, allergic nephritis, and similar conditions, each of which has now been transmitted by sensitized lymphoidal cells (8–11). Adjuvant arthritis differs from each of these conditions, however, since in the latter one or more tissue antigens are added to the initial adjuvant inoculum, whereas none is incorporated prior to injection into our rats. Hence, the identity of the responsible antigen in adjuvant arthritis is still unknown.

Methods

All of the rats used in these experiments were of the highly inbred Lewis strain, and were obtained from Microbiological Associates, Bethesda. Rats of this strain have been used successfully in other transfer experiments and were said to be unable to reject reciprocal skin grafts, although we did not conduct this test. All were healthy females, weighing 150 to 200 gm at the start of the experiment. The animals were kept in roomy cages in groups of six to eight per cage and donors were separated from recipients after they had been given adjuvant inoculations. All were fed on Purina lab chow and water *ad libitum*, and clean conditions were maintained at all times. The adjuvant which was inoculated into donors was composed o 4 parts by volume of a light mineral oil, one part emulsifier (Falba), and 3.2 parts saline to which was added the wax D fraction of the human Canetti strain of the tubercle bacillus[1] in an amount of 1.0 mg/ml of final adjuvant. In order to provide better dispersion of the wax D fraction, it was mixed thoroughly with a small amount of 70 per cent alcohol (usually 0.1 to 0.2 ml to 28 mg of wax) immediately before mixing and emulsifying with the fluid contents of the adjuvant.

[1] Kindly supplied by Dr. E. Lederer, Laboratoire de Chimie Biologique, Faculté des Sciences de l'Université de Paris.

All inoculations into donors were given, as in previously reported experiments (1, 2) into the skin of the superscapular area in multiple small deposit sites, the total amounting to about 0.4 ml in all. In addition, 0.1 to 0.2 ml was administered into one or two foot-pads of some animals. Lymphoidal cells for passive transfer were obtained from the donors in various experiments on from the 4th to the 16th postinoculation days. Cells from the axillary, olecranon, submaxillary, superficial and deep cervical and inguinal nodes were harvested and pooled from several donors (usually 4 to 6 in number). Nodes especially chosen were those in proximity to adjuvant depots and those which could be rapidly extirpated and readily cleaned so that they could be rapidly processed. These procedures assured the harvesting of ample numbers of viable cells. Spleen cells and thymus cells were each pooled separately from multiple donors. The residue or debris which remained after straining out the cells from any of the



TEXT-FIG. 1. General design of a typical experiment.

organ structures was pooled, along with some viable and many dead cells, resuspended in saline, and listed as "sediment."

The cell suspensions were obtained as follows: Lymphoid tissues were removed from living donor rats under ether anesthesia, using antiseptic treatment (70 per cent alcohol) of instruments and the animal's shaved skin before incision. All other apparatus and glassware coming in contact with the excised tissues and the solutions were autoclaved. As fast as tissues were removed from the rat they were placed in a small Petri dish in cooled Hanks' balanced salt solution (HBSS), (12). These were refrigerated immediately at 4°C until sufficient tissues were accumulated for one set of recipient rats. They were then teased apart rapidly with fine forceps in fresh HBSS and pressed gently through a wire sieve (30 apertures per square inch) with a metal spoonula to remove the connective tissue and large pieces which might clog the cytosieve (13). The unstrained debris was stored in a covered Petri dish at 4°C for later intraperitoneal inoculation while the strained material was put through a modified cytosieve and then transferred to a centrifuge tube. HBSS was added to make 12 ml and it

was centrifuged 10 minutes at 1800 RPM. The buffy coat layer was pipetted off into a fresh centrifuge tube having a conical bottom, HBSS added to the 12 ml mark, and after gentle mixing with a pipette, centrifuged at 1400 RPM for 6 minutes. The white cells were again carefully pipetted off into another tube and enough HBSS added to a total volume of 2 or 3 ml. This was the inoculum. It was injected immediately into the recipient animals in a tail vein or otherwise while another operator counted the number of viable leucocytes in the suspension. The inoculation was given usually over a period of 3 to 10 minutes. Tests for the viability of harvested cells were made using the eosin Y dye exclusion test (12) on an aliquot of each pool. Another aliquot was used for cell counting in the standard white blood cell counting chamber.

All recipients were healthy and in the natural state. Splenectomy, irradiation, or other procedure was not carried out beforehand. Each recipient received a specific number of viable cells from either lymph node, spleen, or thymic source. The general outline of the experimental design for many of the experiments is diagramatically illustrated in Text-fig. 1. Occasionally both intravenous and intraperitoneal inoculations were made. The thick sediment was always given intraperitoneally. A few recipients were also given an intracutaneous inoculation of adjuvant on or close to the same day on which the cells were transferred. As a control fresh kidney tissue was pooled from 6 animals on the 11th postadjuvant day, minced in HBSS, and injected intraperitoneally into three recipients. Other investigators (14) have inoculated heat-killed lymphocytes from mycobacterial adjuvant-injected donors into recipients of this same inbred strain of rats.

In all, donor lymphoid, spleen, or thymus cells were transferred from 70 sensitized donors to 28 recipient rats. Five recipients died within a few minutes of the intravenous injection of cells. These were omitted from the series.

RESULTS

An inflammatory polyarthritis was passively transferred from adjuvant-injected or "sensitized" Lewis rats to untreated rats, also of that strain, in a total of 12 out of 23 recipients. These figures probably do not represent the actual incidence that could have been achieved, since as various factors were altered according to information learned from early pilot experiments, it was possible to transfer the disease in later experiments to 5/5 recipients which received more than 2.0×10^8 sensitized viable lymphoid or spleen cells.

Careful adjustment of three factors appeared to be necessary for the successful transfer of arthritis. These included (*a*) the use of at least a certain minimal number of viable lymphoid cells, (*b*) proper selection of the route of administration, and (*c*) selection of a proper time interval after adjuvant injection in the donor before harvesting the sensitized cells.

In the initial pilot experiments, sensitized cells from nodes and spleen were removed on the 10th postadjuvant day and pooled before injection. In this experiment, 0.36 to 1.56×10^8 cells were injected into each of six recipients. None developed arthritis. Subsequently, it was found that the minimum number of viable cells necessary for transfer was 2.0 to 2.3×10^8 if these were obtained from pooling of cells from lymph nodes from various areas of the body. However, in one experiment where cells were taken primarily from the enlarged olecranon nodes, after injection of the adjuvant into the forepaws,

arthritis was transferred with 1.70×10^8 cells, the smallest number that successfully transferred the arthritis. The least number of splenic cells which induced arthritis was 2.70×10^8. On the other hand, it was *not* possible to transfer disease with cells obtained from the thymus gland. Hence, in several trials (total of four recipients) from 4.95×10^8 to 1.19×10^9 thymocytes did not produce any visible disease in the recipients. Thymocytes did not, on the other hand, seem to have an inhibitory effect since one recipient which received a total of 1.12×10^9 mixed node and thymic cells intravenously, as well as receiving some residual node and thymic gland debris intraperitoneally,

TABLE I

Cellular Transfer of Adjuvant Arthritis. Significance of Stage of Donor Preparation and Number of Cells Transferred

Source of cells	Day after adjuvant	No. of cells transferred $\times 10^{8}$*	Arthritis in recipients
Nodes and spleen	5	0.36; 1.91	No, 0/2
Nodes and spleen	7	1.22	No, 0/1
Nodes and spleen	8	1.52	No, 0/1
Nodes and thymus	8	2.81	No, 0/1
Nodes and thymus	9	11.20; 42.63	Yes, 2/2
Nodes	10	1.70; 2.20	Yes, 2/2
Spleen	10	1.78	Yes, 1/1
Spleen	10	1.78	No, 0/1
Nodes and thymus	11	0.39	No, 0/1
Nodes	11	3.82; 8.05	Yes, 2/2
Spleen	11	2.70; 3.83; 3.83	Yes, 3/3
Thymus	11	11.96; 11.96	No, 0/2
Nodes or thymus or spleen	14	2.10 to 4.95	No, 0/4

* Cells given intravenously to each recipient.

developed significant polyarthritis 7 days later. A summation of most of the transfer experiments is given in Table I.

The intravenous route for administration of cells to recipients seemed to be by far the most satisfactory method and was used in nearly all of the experiments. When intraperitoneal or subcutaneous routes were used solely in five recipients, no disease developed despite the number of cells injected or their source, except in one animal which showed a minimal swelling of one heel for 6 days after intraperitoneal injection of 4.26×10^9 mixed node and thymic cells. Six additional animals which received an intracutaneous injection of adjuvant on or within 1 or 2 days of the time of transfer of sensitized donor cells did not differ materially in their ability to develop arthritis, either in intensity or time, from those which only received the cell suspension. Because of the complexities that

could arise from inclusion of these animals in the final evaluation they were excluded from further consideration in the series.

Although sufficient studies have not been conducted in order to draw unequivocal conclusions about this point, the impression has been gained that

TABLE II

Transfer of Adjuvant Arthritis by Cells Obtained from Rats Sensitized 10 and 11 Days Previously

Donors				Recipients							
Rat No.	Day of onset of arthritis	Severity score on day 11*		Rat No.	Source of cells	No. of viable cells × 10^6	Route of injection	Arthritis	Onset day	Highest score/day	Duration
											days
Experiment 1-63-J-N											
1	10	5		A.	Nodes and sediment,‡ 1.5 ml	3.82	i.v. i.p.	Yes	5	4/10	22
2	10	8	Cells harvested on day 11	B.	Spleen and sediment,‡ 1.5 ml	2.70	i.v. i.p.	Yes	3	6/14	15
3	11	2		C.	Thymus	11.96	i.v.	No	—		
4	10	4		D.	Thymus	11.96	i.p.	No	—		
5	10	5		E.	Sediment, 4.0 ml	—	i.p.	No	—		
Experiment 3-63-B											
6	10	1		F.	Nodes (olecranon)	1.78	i.v.	Yes	3	3/5§	—
7	10	3	Cells harvested on day 10	G.	Nodes	2.20	i.v.	Yes	3	2/5§	—
8	10	2		H.	Spleen	1.78	i.v.	Yes	4	1/2	6
9	10	1		I.	Spleen	1.78	i.v.	No	—	—	—
10	10	1									
11	—	0									

* Maximum score is 20 for the severest degree of arthritis (1).

‡ Sediment = debris remaining after removal of viable cells from nodes, spleen, and thymus.

§ These recipients were sacrificed on day 5 for histological study.

there is a critical time period after adjuvant, during which lymph node or spleen cells have the capacity to transfer arthritis to normal recipients. Hence, cells for all of the successful transfer experiments were obtained between days 9 and 11, during which time a polyarthritis was just developing in the donors. Two representative experiments are detailed in Table II where it will be noted that an arthritis of mild or moderate degree had already developed in 5/5 and

5/6 donor animals 1 or 2 days previously. Earlier transfer experiments made on days 5, 6, 8, or 9 were unsuccessful but these experiments were parts of the early study when less than 1.56×10^8 cells were given to each recipient. On the other hand, in one later experiment, ample numbers of lymph node, spleen, or thymus cells were harvested on day 14 from five donor rats and were given intravenously or intraperitoneally to four recipients. None of them developed disease (Table III). It is interesting to note in the donor animals that the lymph

TABLE III

Failure of Transfer of Adjuvant Arthritis by Cells Obtained from Rats Sensitized 14 Days Previously

Donors				Recipients				
Rat No.	Onset day	Severity score* on day 14		Rat No.	Source of cells	No. of viable cells $\times 10^8$	Route of injection	Arthritis
Experiment 1-63-J-O								
6	11	9	Cells§ harvested on day 14	F.	Nodes and sediment,‡ 0.8 ml	2.10 —	i.v. i.p.	No
7	11	9		G.	Spleen and sediment,‡ 1.5 ml	2.90 —	i.v. i.p.	No
8	10	10		H.	Thymus and sediment, 1.5 ml	4.95 —	i.v. i.p.	No
9	10	9		I.	Sediment, 4.0 ml	—	i.p.	No
10	10	12						

* Maximum score possible is 20 for the severest degree of arthritis.

‡ Sediment = debris remaining after removal of viable cells from nodes, spleen, and thymus.

§ Nodes reduced in size, flabby, with nodular pearly hard centers. Thymuses average size or small. Spleens enlarged and containing white granulomas.

nodes and thymus gland were small, flabby, and appeared to be fibrotic and depleted of many of their cellular constituents. By the time that the lymph node and other cells were harvested from these five donor animals, all of them had developed a florid and severe polyarthritis. It seems quite possible that most of the sensitized cells had by this time been released to the joint tissues where they were involved in the inflammatory articular disease.

By contrast, the lymph nodes removed for cell harvesting on days 8 to 11 were enlarged and often quite firm. This was especially the case with the axillary and cervical nodes when adjuvant had been given into the skin of the superscapular region, or with the homolateral olecranon and axillary nodes when

adjuvant was given into a forepaw. The olecranon nodes were often enlarged two to three times their normal size, and were usually pink and quite firm. A few contained small white spots, suggestive of adjuvant deposition. As some of these nodes were dissected, small droplets of oil, obviously embolized adjuvant, floated out. As much as possible of this material was removed by repeated washing and gentle centrifugation.

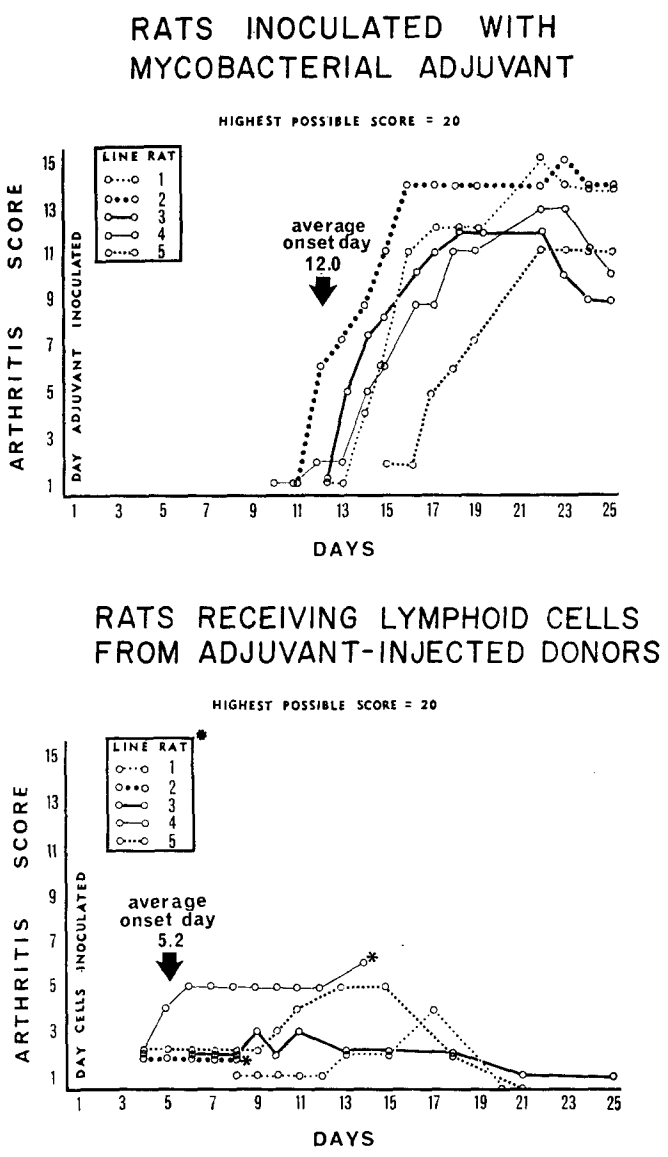
The spleens of donors were slightly enlarged and moderately congested. Occasionally one or more greyish-white spots 0.5 to 2.0 mm in diameter were present in the subcapsular zone. This was especially notable in spleens of 14-day donors. Histologically, these were small reactive granulomas surrounding embolized adjuvant material. The thymuses varied considerably in size. In general, those that were removed on the 7th to 10th postadjuvant inoculation days were somewhat enlarged, pink, and sometimes contained a few hemorrhagic spots. On the 12th and 15th days, the thymuses were either normal in size or smaller than normal.

In Lewis rats, as in other strains previously studied, injection of adjuvant was invariably followed by a 9 to 12 day latency period prior to the onset of polyarthritis. In several hundred animals, the disease has never occurred before the 9th day and usually does not appear until the 11th or 12th day (Text-fig. 2). These observations are of special importance since onset of the transferred disease usually occurred on the 3rd to 6th days *after* inoculation of sensitized cells (Text-fig. 2). In the 12 recipients which developed lesions, 7 did so on day 3 or 4, 1 on day 2, and 4 on days 5, 6, 7, and 8 respectively (over-all average, 4.3 days).

The lesions which developed in the recipients were identical in the gross in type and form to those observed after adjuvant inoculation (1, 2) but generally they were not as severe or extensive (Text-fig. 2). Based upon a rating system of 20 for over-all maximum arthritis (1), 15 adjuvant-injected Lewis control animals had an average maximal score of 7.7 whereas the transferred arthritis in 12 animals had an average maximal score of 2.3. Also, the actively induced arthritis was of longer duration than the transferred disease.

Periarticular swelling and erythema developed in the passively sensitized animals over the metacarpal phalangeal, metatarsal phalangeal, and proximal interphalangeal joints (Fig. 1), along the tarsal bones, about the ankles and wrists, and occasionally in the tail between the caudal vertebrae. Cutaneous and subcutaneous erythematous nodules also appeared on the dorsum of either hind- or forepaws (Fig. 2), on the tail, or along the course of the vessels in the ears (Fig. 3). All of these findings are similar to those which have been noted in animals actively sensitized with adjuvant, (1, 2, 15), although in the latter, the lesions were more extensive and the periarticular swelling and erythema were usually much more marked.

Histologically, the fully developed lesions were also similar to or identical



*Rats 1-4 received node cells; rat 5 received spleen cells.
*Rats killed for histological study.

TEXT-FIG. 2. Comparison of latency period and severity of arthritis of actively and passively sensitized animals.

with those observed in actively sensitized rats (16, 17). The passively induced lesions consisted of (*a*) inflammatory synovitis and subsynovial edema (Figs. 4 and 5), (*b*) proliferation of connective tissue elements in focal areas in the subsynovium, subcutaneous regions of the paws and ears, peritendonous zones, and periosteum, (*c*) periosteal new bone formation, (*d*) some early granulation tissue (pannus) formation in a few animals, and (*e*) inflammation and connective tissue reactions adjacent to the intervertebral discs between the vertebrae of the tail (Fig. 6). The inflammatory cellular elements consisted predominantly of pleomorphic mononuclears, many of which resembled large lymphocytes or histiocytes (Fig. 7). Interspersed were also a few small lymphocytes and occasional plasma cells or neutrophils. Some inflammatory foci were clustered in the immediate subsynovial regions as shown in Figs. 4 and 5. Usually large numbers of proliferative connective tissue elements resembling primitive fibroblasts were interspersed in and about these inflammatory cells.

The histology of the subcutaneous nodules in the external ears was identical with that described in active adjuvant arthritis (16, 17). It consisted of perivascular (perivenous) accumulations of mononuclears, some localized edema, hyperemia, and occasionally acanthosis of the adjacent epidermis with microabscess formation.

The histology of the passively transferred arthritis has only been studied from the time of clinical recognition of lesions through the 20th day after its clinical appearance. In the latter case, a moderate degree of continuing inflammation and connective tissue activity was present. It is likely that cellular invasion begins several hours or 1 or 2 days before it becomes clinically evident, but we have no evidence on this point. Furthermore, the limits of persistence of the gross or microscopic disease following a single injection of sensitized cells, or repeated administration of them, is not known at this time.

DISCUSSION

Adjuvant arthritis has been considered to be an immunologically induced disease, but most of the evidence in this respect has been indirect. Thus, the latency period of approximately 10 days after adjuvant injection, the positive correlation between the severity of the reaction at the local injection site and the subsequent development of arthritis, the ability to induce immune tolerance-like phenomena, and the inhibitory effects of whole body irradiation and corticosteroids (2, 18) all favor this conclusion and the inability to culture an infectious agent (3, 19) has removed from active consideration another etiological possibility. The strongest evidence so far adduced for an immunologic etiology of adjuvant arthritis is the passive transfer of this condition as reported herein.

Very recently Waksman and Wennersten (14) have also described success

in passive transfer of adjuvant arthritis in rats using essentially the same technique and in fact the same Lewis strain of inbred rats, except that they splenectomized nearly all donor rats 3 to 4 weeks prior to use for transfer of cells. We did not find this maneuver to be necessary, although it may have been responsible for the somewhat more severe passive disease which developed in Waksman and Wennersten's animals. The latency period of 2 to 8 days, average, 4.3 days in our series, is much shorter than the average (11.3 days) in a large series of actively sensitized animals of either the Lewis strain or others with which we have dealt. Negative controls in our series consisted of animals injected with (*a*) sensitized cells disrupted by repeated freezing and thawing, (*b*) "debris" composed of a few viable and many non-viable cells and stroma from either nodes, spleen, or thymus, or (*c*) homogenates of kidney tissue. Waksman and Wennersten could not passively transfer disease after gentle heating of suspensions of sensitized cells or by transfer of control cells from donors injected with oil alone. These results point to the specificity of the reaction and the need for viable lymphoid cells, thus indicating the immunologic character of adjuvant arthritis.

Of added interest were two other points. The first was that, although not proven conclusively, it appeared that lymph node or spleen cells were not competent to transfer the disease until approximately the 8th day after adjuvant injection. Thereafter, sufficient numbers of sensitized cells were present in these tissues only until about the 11th postadjuvant day after which the nodes and, to a lesser degree, the spleen appeared to have released large numbers of cells which probably localized in and about the joints and produced the clinical disease. The nodes at this time were small, flabby, and obviously much less replete with cells than they were several days previously. On the 14th postadjuvant day, it was not possible to transfer a clinical disease, even when large numbers of cells were pooled and transferred. Presumably by this time most or all of sensitized cells had been discharged from these tissues and the cells which were harvested were either a fresh non-sensitized crop or were comprised chiefly of stromal and other non-sensitized types. A second interesting observation was that cells obtained from the thymus gland did not have the capacity to transfer adjuvant arthritis regardless of when after adjuvant injection they were obtained.

The clinically recognizable passively transferred lesions which are produced in and about the joints and in the subcutaneous tissues are similar in all respects, except perhaps in severity, to those noted in classical adjuvant arthritis. Probably the ability to develop such detectable lesions is related both to the time after adjuvant injection when the donor cells are harvested, and the number of sensitized cells which are given. It is likely, although as yet unproved, that even small numbers of cells can produce histologically detectable lesions in and

about joints and adjacent tissues, whereas larger numbers of sensitized cells or repeated injection of such cells given at appropriately spaced intervals could cause a much more severe or a chronic and perpetuated disease.

The localization of lesions in the synovium and adjacent connective tissue, and in the subcutaneous regions of certain parts of the body signifies that a degree of tissue specificity exists in the sensitized cells. Visceral organs rarely contain any lesions or cellular collections in actively sensitized animals, except for occasional granulomas in the lung and spleen in some animals (1). None were observed in the passively sensitized recipients who developed arthritis.

Is the small amount of adjuvant which must have been unavoidably transferred along with the node or spleen cells sufficient to induce an active arthritis in the recipient animals? It probably is not for the following reasons: (*a*) it is known that a minimal quantity of about 0.04 ml of adjuvant containing tubercle products is required in order to induce active disease (20). It is not possible that such an amount would be transferred in the repeatedly washed cell suspensions, since although some adjuvant is known to reach regional nodes and spleen soon after inoculation (20) and some also migrates to the paws and lungs, the great majority (probably 50 to 70 per cent) remains for several days or weeks in the local injection site; (*b*) the appearance of arthritis 3 or 4 days after cell transfer renders any kind of actively induced arthritis most unlikely, since regardless of the adjuvant dose, whether it be large or small, or the route of administration, it has never been possible to induce active disease sooner than 9 days after inoculation.

The two pressing questions that this study does not answer concern the nature of the antigen and the precise mechanisms involved in the development of the disease process. Since adjuvant arthritis can now be considered with reasonable assurance to be caused by some type of immunological reaction, a more directive effort can be made to answer these questions.

SUMMARY

An adjuvant-induced arthritis has been passively transferred in a highly inbred strain of rats by transfer of viable lymph node or spleen cells, but not thymus cells, to normal recipients. After an interval averaging 4.3 days recipients developed arthritis, whereas animals actively sensitized with adjuvant never developed disease before the 9th day (average 11.3 days). The transferred disease had all of the gross and pathological characteristics of primary disease, except for a lesser severity. Control studies using non-viable cells either of lymphoidal or other tissue origin were always negative. It is concluded that adjuvant arthritis is the result of an immunologic reaction which is perhaps similar to delayed hypersensitivity. The antigen in this reaction so far remains obscure.

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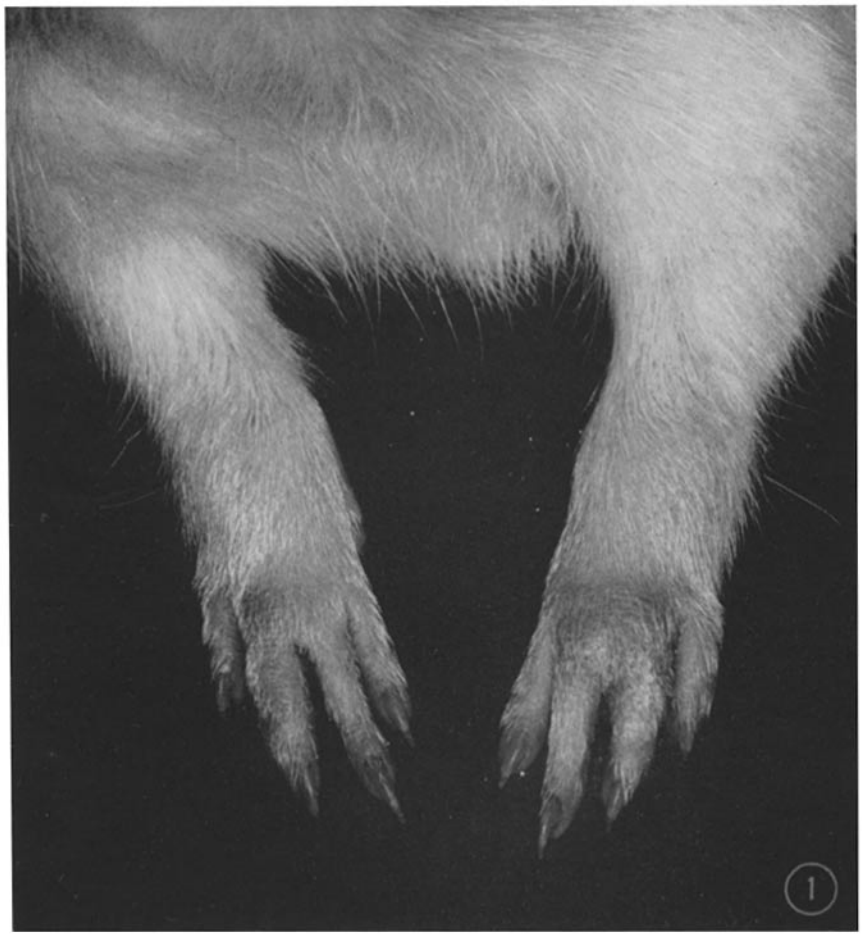
EXPLANATION OF PLATES

PLATE 42

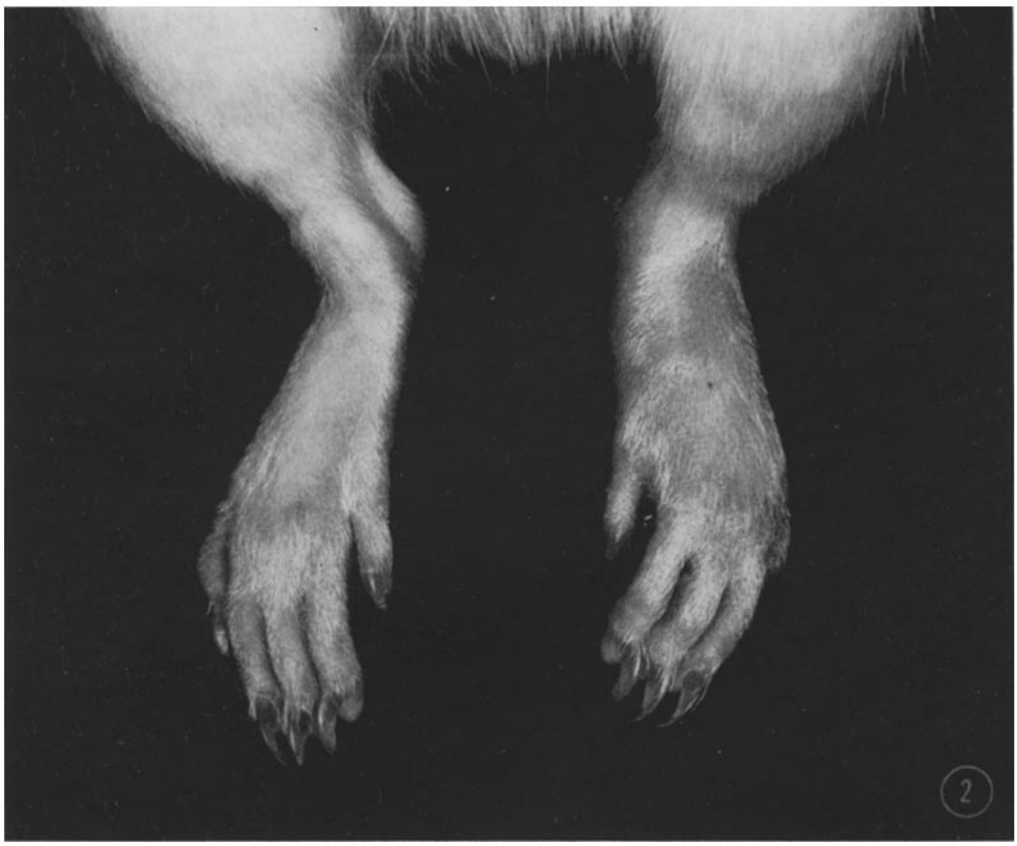
FIG. 1. Passively transferred arthritis in both forepaws as seen 10 days after transfer of lymph node cells. Onset of arthritis occurred in the 4th post-transfer day. Adjuvant was also given to this animal on the day the cells were inoculated.



(Pearson and Wood: Passive transfer of adjuvant arthritis)

PLATE 43

FIG. 2. Passively induced arthritis 4 days after transfer of 4.52×10^8 lymphoid cells. Note diffuse and mottled swelling of left hindpaw and 2 or 3 small erythematous areas on the right paw.



(Pearson and Wood: Passive transfer of adjuvant arthritis)

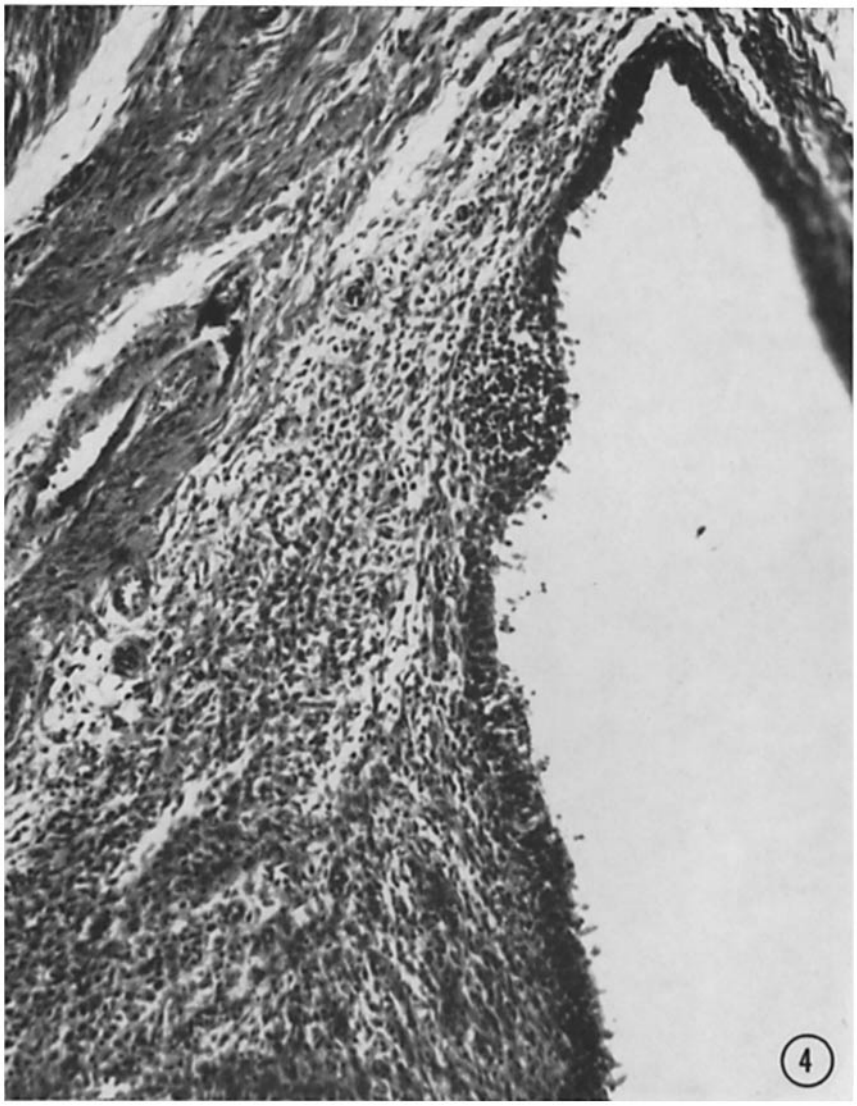
PLATE 44

FIG. 3. A group of five erythematous perivascular lesions are seen in the external ear in this animal 7 days after receipt of large numbers of lymph node, spleen, and thymus cells.

(Pearson and Wood: Passive transfer of adjuvant arthritis)

PLATE 45

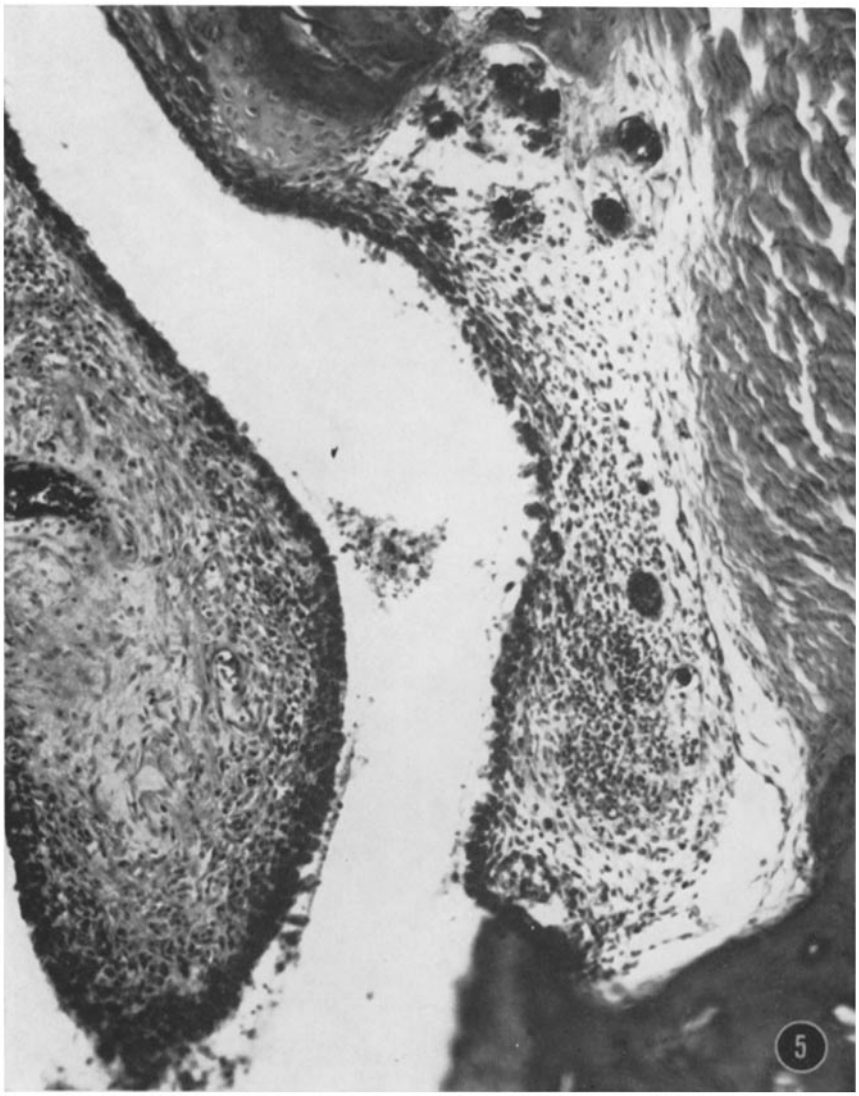
FIG. 4. Recipient 10 days after transfer of sensitized cells and 6 days after onset of arthritis. Metatarsal joint showing intense inflammation, edema, synovial proliferation, and an immediate subsynovial cellular collection. Hematoxylin and eosin. × 162



(Pearson and Wood: Passive transfer of adjuvant arthritis)

PLATE 46

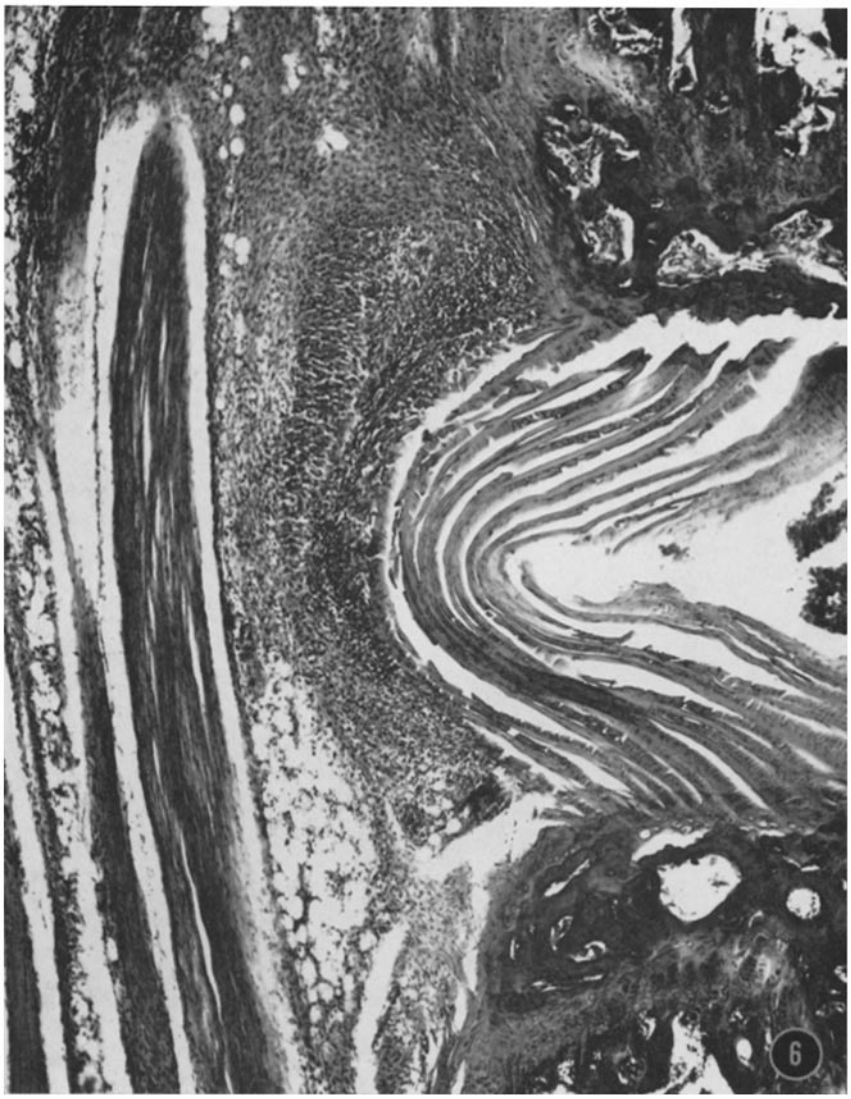
FIG. 5. Recipient as in Fig. 4. Tarsal joint showing cellular reaction and vascular congestion in a fibrous villus and adjacent synovium. Hematoxylin and eosin. × 162.



(Pearson and Wood: Passive transfer of adjuvant arthritis)

PLATE 47

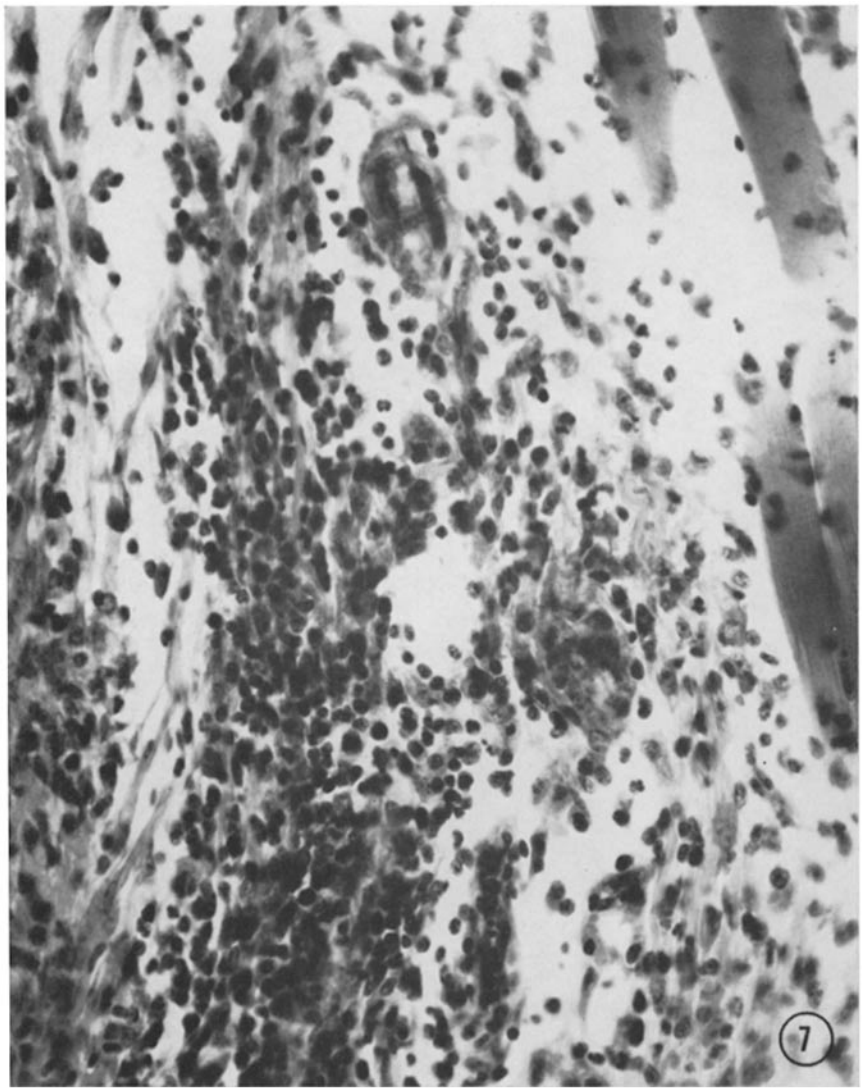
FIG. 6. Recipient as in Fig. 4. Tail. Intense inflammation adjacent to a caudal intervertebral disc. Hematoxylin and eosin. × 50.



(Pearson and Wood: Passive transfer of adjuvant arthritis)

PLATE 48

FIG. 7. Recipient as in Fig. 4. Higher magnification to illustrate the types of inflammatory cells which are involved in the reactive response. Hematoxylin and eosin. × 410.



(Pearson and Wood: Passive transfer of adjuvant arthritis)