stronger avidity than the uncommitted cells[24,25]. Assuming that the differentiation of uncommitted precursor cells, as well as the occurrence of antibody-producing cells, distinctly depends on the dose used for primary antigenic stimulation[26], and making the additional assumption that the passively administered 7S antibody is capable of inhibiting the build-up of immunological memory up to a certain degree of the differentiative processes of uncommitted progenitor cells, one may suggest that priming with tiny doses of antigens, such as 5×10^5 SE, leads to the formation of memory cells still carrying antibody-analogous receptors with relatively low avidity. Under those prerequisites one might expect that the specific 7S antibody passively administered together with a booster dose of 10^8 SE is capable of producing effective suppression of the secondary immune response. The findings obtained (figure 2) support this concept. Such experiments with tiny doses of antigens as a primary antigen stimulus are evidently very similar to the natural conditions of Rh-negative women at risk. The question arising which volume of fetal blood with Rh-positive erythrocytes will effect sensitization of Rh-negative women has not been answered by different authors in the same way[27]. This is not surprising, since it has been learned more recently that the immunogenicity of the D antigen of human erythrocytes from different donors will show considerable variance in human volunteers[28,29]. Taking into consideration that on one hand the individual sensitization risk of a Rh-negative nullipara by her Rh-positive child depends on the number of fetal erythrocytes present in maternal circulation immediately after delivery[30], and that on the other hand there exists no method for an exact determination of the volume of transfused foetal blood[13,31], it appears, on the basis of the experimental findings reported here, to be still worthwhile to administer the anti-Rh even if delivery has occurred more than 72 h previously.

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Experimental allergic encephalomyelitis (EAE)-changes in structure and proliferation in rat lymph nodes after sensitization with guinea-pig and bovine basic myelin protein

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Summary. Substantial difference in the proliferation of lymphoid cells in the draining LN was found in rats injected with guinea-pig EBP-FCA and bovine NBP-FCA indicating significance of the encephalitogenic determinant in the myelin basic protein in the peripheral lymphatic reaction initiating EAE.

Regional lymph nodes (LN) draining the site of injection of the encephalitogenic antigen [basic myelin protein (EBP) mixed with complete Freund's adjuvant (FCA)] play a key role in the development of EAE. If such nodes are removed within 5 days after sensitization, no EAE develops[1]. It has been shown that the dynamics of growth of LN, as well as their histological structure, after administration of an EBP-FCA mixture differ markedly from changes produced by FCA alone[3]. The present work involves an attempt to specify the antigenic action of EBP on peripheral lymphatic reactions. To this end we have compared changes in structure and proliferation kinetics of regional LN after administration of encephalitogenic guinea-pig basic protein (EBP-FCA) and bovine basic protein (NBP-FCA) which does not induce EAE in the rat.

Cell proliferation in regional lymph node after injection of EBP-FCA, NBP-FCA and FCA alone, as compared with controls injected with saline

	EBP-FCA	NBP-FCA	FCA	Controls
Experiment I	11.3 ± 1.27*	5.5 ± 0.81	–	1.2 ± 0.10
Experiment II	10.9 ± 0.20	6.6 ± 1.05	5.8 ± 0.69	1.5 ± 0.01

*LI (%) ± SEM.

Material and methods. Female rats of the inbred Lewis strain, aged 10 weeks, were used. The EBP was prepared from guinea-pig brain by the method of Eylar[2]. The NBP was prepared from bovine material by the same method. Both antigens were injected intradermally into the left hind footpad in a dose of 400 µg per animal, dissolved in 0.1 ml saline and emulsified in the same volume of FCA (olive oil : lanolin : Tween 80, 15 : 4 : 1 and heat-killed Mycobacterium tuberculosis, 8 mg/ml adjuvans). Some animals received only FCA. Controls were injected with 0.2 ml of saline. On the 4th day after sensitization, the animals received i.p. 1 µCi 6-^{3}H thymidine/g b.wt (ÚVVVR, Prague, spec. act. 19–20 Ci/mMol); 60 min after injection, the lymph nodes were removed and processed as described elsewhere[3]. 2 and 7 µm thick slices were cut from the Paraplast-embedded material; 7 µm thick slices were stained with Mayer's hematoxylin and eosin. Autoradiograms were prepared from 2 µm thick slices as described elsewhere[3]. From each animal 2000–3000 cells were counted to determine the labelling index (L.I.).

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Results. The EBP-FCA caused accumulation of lymphocytes and large lymphoid cells in the paracortex and in the medullary cords which was more marked than after injecting NBP-FCA or FCA alone. The development and amount of granulomatous tissue in the hilar area was more prominent in the EBP-FCA and NBP-FCA groups. In the EBP-FCA group, the enlargement of the paracortex was especially prominent; the traffic of lymphocytes through the high endothelium venules was very intensive, and a high proportion of cells of the plasmocytic series were present in the medullary cords which almost compressed the sinuses. Autoradiographic estimates of cellular proliferative activity in the nodes showed that number of proliferating cells significantly increased in both experimental groups. In the control group, L.I. ranged from 1.2% to 1.5%, in the NBP-FCA group from 5.5% to 6.6% and in the EBP-FCA group it was approximately 11%; in the FCA group L.I. was 5.8% (table).

Discussion. Previous work[3] has shown that peak LN wt increment involves a growth of the lymphoid cell population mainly within the first 4 days. Following day 4, the growth of non-specific granuloma prevails. For this reason we concentrated our observations in the present work on the early stages of disease induction. According to previous findings[3], LN growth after NBP-FCA was significantly slower than after EBP-FCA in the same time interval. Present results revealed that EBP-FCA resulted in a more intense accumulation of lymphoid cells in the paracortex and a greater development of granuloma than NBP-FCA or FCA alone. Maturation of plasma cells was also accelerated after EBP-FCA. NBP-FCA, as compared with FCA alone, increased somewhat the growth of the lymphoid cell population and the medullary cords. The significance of the latter must, however, be interpreted with care, since compression from the FCA developing lipogranuloma substantially changes the circulation and architecture of the LN. The latter might be expressed more clearly with EBP and NBP without the addition of FCA. Eliciting EAE without FCA is, however, difficult, if at all possible. Autoradiographic data indicate that the nodal growth in both experimental groups involved proliferation of cells in situ. Previous report[3] showed that growth of the cell population involves also migration of lymphocytes into the LN. Following NBP-FCA injection, both processes are therefore apparently less intense and approximate values for administration of FCA alone. The differences in LN growth and proliferation observed after administration of EBP-FCA and NBP-FCA show that non-specific stimulation of LN by basic protein without encephalitogenic determinant (ED) is relatively small. The presence of ED in the basic protein molecule (EBP-FCA) markedly influenced the local reaction of the LN draining the site of injection in terms of increasing migration of cells into the nodes and proliferation of cells in situ.

Relation between stimulus intensity and neutrophil chemotactic response[1]

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Summary. The effect of chemotactic peptides which lack chemokinetic activity has been investigated. The neutrophil response is proportional to the logarithm of the stimulus intensity, or alternatively a power function with an exponent of 0.3. Equal responses are obtained for equal ratios between the peptide concentration in the lower compartment and the threshold concentration. The significance of Weber-Fechner's law in leucocyte chemotaxis is discussed.

The direction of actively moving leucocytes can be determined by chemical substances in the environment. Since Leber's studies in 1888[3], it is generally believed that this process, called chemotaxis, is instrumental in leucocyte accumulation at inflammatory sites. The relationship between the intensity of the chemotactic signal and the direction finding of leucocytes as expressed in directional locomotion is still largely unknown. The interpretation of earlier data[4–6] was complicated by the fact that the test material influenced the speed of the cells, as well as their direction of locomotion. Thus it had chemokinetic as well as chemotactic activity. It was therefore necessary to evaluate the relationship between stimulus intensity and directionality by means of purified cytotaxin preparations exhibiting chemotactic activity only. Partially purified peptide preparations containing classical anaphylatoxin (S-CAT 1.5.1)[7,8] were prepared from dextran-activated swine serum for such experiments. This preparation contained 4% of chemotactically active peptides[9]. Random and directional locomotion of human peripheral blood neutrophils was assessed with a modified filter technique[10], which provides for stable gradients (unpublished observations). The behaviour of responding cells (random vs directional locomotion) was also determined by direct observation. The results obtained with these 2 techniques were in agreement. Human serum albumin (HSA) had to be present in the test system to permit efficient movement of cells and thereby the expression of chemotaxis in form of directional locomotion[11].

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