inhibée par l'acétylsalicylate de lysine pourrait confirmer l'hypothèse selon laquelle l'aspirine agirait sur l'aggrégation des plaquettes par son activité «anticholinestérasique».

Conclusion. En utilisant une forme soluble d'aspirine, l'acétylsalicylate de lysine, nous avons pu mettre en évidence chez le rat son action inhibitrice sur les pseudocholinestérases des parois des vaisseaux du cerveau. Ces résultats pourraient rendre compte de l'activité de l'aspirine dans l'aggrégation plaquettaire.

Summary. Using an injectable solution of acetyl salicylate lysine salt it was shown that aspirine has an action on the pseudocholinesterases in the brain vessels of the rat. This could help to explain the activity of aspirine in case of platelets aggregation.

B. DELBARRE, G. DELBARRE et P. JOBARD[11]

Laboratoire d'Anatomie pathologique et Clinique Médicale de la Faculté de Médecine, 2bis, Boulevard Tonnellé, F-37 Tours (France), 11 février 1971.

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The Effects of Sodium Salicylate on the Anamnestic Immune Response in vitro

Divergent results have been reported concerning the effect of salicylates on the antibody response in vivo[1]. Recently, a suppressive effect in vivo on the number of spleen antibody-forming cells was demonstrated in mice[2]. AMBROSE[3] showed that salicylates, when added during the 'inductive' period, inhibited the in vitro anamnestic immune response in rabbit lymph node fragments.

Salicylates (30/mg 100 ml) inhibited the antigen and phytohemagglutinin-induced increase of the synthesis of DNA, RNA and protein in human blood lymphocytes in vitro[4,5].

In the present communication, we have studied the relationship between this salicylate inhibition of cellular DNA synthesis and the synthesis and release of IgM and IgG antibody using the anamnestic response of immune chicken spleen cells to sheep erythrocytes (SRBC) in vitro.

Eight-week-old white leghorn chickens were immunized i.v. with 5×10^9 sheep erythrocytes (SRBC) per kg body wt. They were sacrified 8 days later and spleen cell suspensions were prepared and cultured as described elsewhere[6]. In brief, tightly stoppered tube cultures containing 15×10^6 spleen cells in 2 ml Waymouth's MB 752/1 medium supplemented with penicillin (100 U/ml), streptomycin (100 µg/ml) and homologous chicken serum (15%) were used. The gas phase was 5% CO_2-95% air and the culture temperature 37.5 °C. Half of the cultures received 10^7 SRBC each.

The incorporation of H^3-methyl-thymidine (H^3-TdR), indicating the DNA synthesis of the cultures, was determined daily as previously described[7]. Antibody-producing, plaque-forming cells (PFCs) in the cultures were determined by the direct and the indirect versions of the Jerne technique[8] as previously described[7].

Sodium salicylate (Matheson, Coleman and Bell, Cincinnati, Ohio, USA) was added to the cultures to a final concentration of 34.8 mg/100 ml (2.17 m*M*). The effects of the drug during a 5-day-culture period is illustrated by one typical experiment; 5 experiments were performed in all.

Figure 1 demonstrates the effects of salicylate on the H^3-TdR incorporation in cultures with and without SRBC antigen. Cultures with antigen were more inhibited than cultures without antigen. Salicylate decreased the magnitude of the H^3-TdR incorporation, while the rate of increase, which was approximately logarithmic, was unchanged.

Figure 2 shows the antigen-induced increase in the number of direct PFCs (IgM) and indirect PFCs (antibodies of low hemolytic efficiency, presumably mostly IgG). Cultures without antigen contained practically no PFCs. The number of direct PFCs increased, with time, in approximately logarithmic progression. Salicylate strikingly decreased the number of direct PFCs (proportionately more than the H^3-TdR incorporation) but the

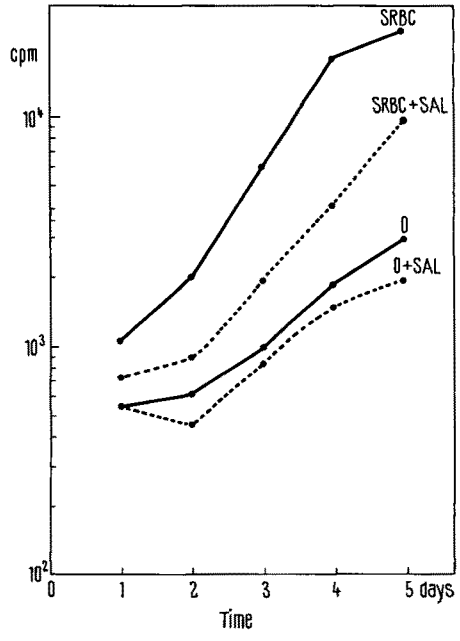

Fig. 1. The effect of sodium salicylate (SAL) on the H^3-TdR incorporation (counts per min) in cultures with and without SRBC.

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The effect of sodium salicylate on the H^3-TdR incorporation in cultures with and without SRBC and on the number of direct and indirect PFCs. Mean suppression (%) ± S.E.M. in 5 experiments

Measured parameter	Day of culture				
	1	2	3	4	5
H^3-TdR SRBC	27± 2	54± 2	62± 2	67± 5	45±16
— H^3-TdR	10± 4	13± 7	26± 7	27± 5	−3±18
Direct PFCs	−34±76	79±13	93± 3	95± 2	86± 7
Indirect PFCs	6± 8	1±17	−17±37	56±10	60± 5

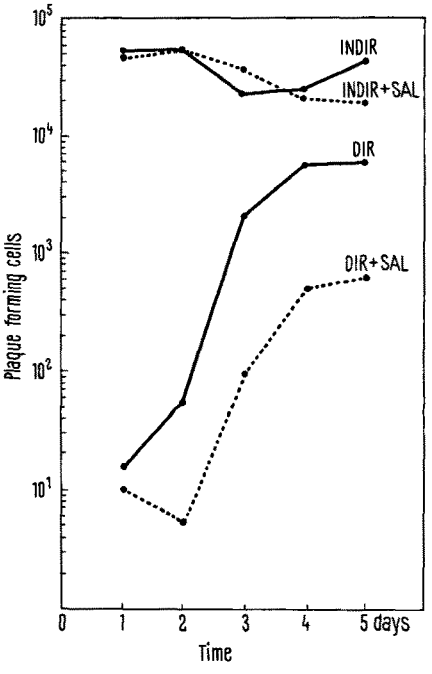

Fig. 2. The effect of sodium salicylate (SAL) on the number of direct (DIR) and indirect (INDIR) PFCs in cultures with SRBC.

rate of the increase was unaffected. In contrast the number of indirect PFCs (IgG) was much less affected by sodium salicylate. A suppressive effect was found on the last 2 days of the culture period.

The Table summarizes the effects of salicylate, expressed as per cent suppression, on the in vitro immune response to SRBC. The results indicate in general that sodium salicylate is an inhibitor of the in vitro anamnestic immune response. The drug suppressed the number of direct PFCs > antigen-induced H^3-TdR incorporation > the number of indirect PFCs > background H^3-TdR incorporation. This indicates a certain selectivity in its action.

Phytohemagglutinin-stimulated lymphocytes preparing for mitosis show an increased dependency on mitochondrial function[9]. Therefore the action of the salicylate may be a result of its uncoupling effect on oxidative phosphorylation and subsequent inhibition of the energy-consuming processes of proliferation and antibody synthesis. In addition, we habe been unable to demonstrate a direct inhibition of the secretion of preformed antibody from spleen cells incubated for up to 6 h with sodium salicylate (ALM and PACHMAN, unpublished results)[10].

Zusammenfassung. Na-Salicylat bewirkt in vitro eine Hemmung der sekundären Immunreaktion von Hühnchenmilzzellen gegen Schaferythrozyten.

G. V. ALM[11] and L. M. PACHMAN

The Variety Club Research Center of the La Rabida-University of Chicago Institute and The Department of Pediatrics, University of Chicago, Chicago (Illinois 60649, USA), 15 February 1971.

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[11] Address: Department of Histology, University of Uppsala, V. Agatan 26 A, S-75220 Uppsala (Sweden).

Nucleolar Alterations of Peripheral Nerve Cells in Rats Following Administration of 4-Hydroxyaminoquinoline 1-Oxide

Neuronal injuries have been noted to occur in animals following administration of certain carcinogenic or carcinostatic agents. McDONALD et al.[1] have demonstrated that brains of mice receiving an injection of nitrogen mustard exhibit neuronal shrinkage in the neocortex, pyriform cortex, hippocampus, cerebellum and medulla oblongata. Administration of cycasin to young mice has been reported to induce ataxia[2] associated with necrosis of granular cells of the cerebellum[3]. KOENIG et al.[4] have shown nucleolar-cap formation of the anterior motoneurones in cats given an intraperitoneal injection of actinomycin D. In the present study, we also found that an i.v. injection of 4-hydroxyaminoquinoline 1-oxide, a compound known as a potent carcinogen, produced nucleolar segregation of the peripheral nerve cells in rats.

Materials and methods. 40 mg of 4-hydroxyaminoquinoline 1-oxide hydrochloride (4HAQO × HCl) was dissolved in 1.0 ml of 0.1 *N* HCl and diluted to a volume of 20 ml with physiological saline. 24 five-week-old male Sprague-Dawley rats were given an i.v. injection of this solution into the tailvein in a dose of 10 mg of 4HAQO × HCl per kg body wt., and 4 rats each were then sacrificed 30 min, 2, 6, 18, 48 and 72 h after injection. As the control group, 5 rats were i.v. injected with 0.5 ml of 0.005 *N* HCl and sacrificed after 6 h. These animals were perfused through the ascending aorta with cacodylate-buffered 3% glutaraldehyde for 30 min at a pressure of 3 ft water under Nembutal anesthesia. After completion of the perfusion, the ganglia of L_4 and L_5, and trigeminal ganglia were excised. In some rats, the coeliac ganglia, supracervical ganglia and terminal ileum were also excised. Tissue samples were fixed again in cacodylate-buffered 3% glutaraldehyde for 30 min, post-fixed in cacodylate-buffered 2% osmium tetraoxide for 45 min, dehydrated through a series of alcoholic concentrations, and embedded in Epon 812. Sections were cut on an LKB ultramicrotome, stained with uranium acetate-lead citrate, and examined with a JEM 6C electron microscope.

Results and discussion. In agreement with the findings by several authors[5,6], neuronal cells of the spinal and trigeminal ganglia from control rats were shown to possess large, irregulary contoured nucleoli consisting of granules, fibrils and amorphous materials. The granules were uniformly distributed constituting more than half of the total