PN 1260

On a biochemically special form of infantile amaurotic idiocy

Infantile amaurotic idiocy (Tay-Sachs' disease) is a genetically dependent, human "inborn error of metabolism" which is caused by a storage of gangliosides within the ganglionic cells of the central nervous system. The gangliosides were discovered by KLENK[1] in the brain tissue obtained from Tay-Sachs' cases. He did considerable work in characterizing this family of substances. Later, they were recognized as normal constituents of nerve cells. In 1959, SVENNERHOLM[2] found that up to 90% of the gangliosides stored in this pathological condition are composed of a particular ganglioside which normally accounts for less then 1% of the gangliosides in the brains of children[5].

The gangliosides are sphingolipids. The lipophilic part of the molecule is an amide (called "ceramide") of a higher fatty acid (in the brain predominantly stearic acid) with the fatty amino alcohol sphingosine. The hydrophilic part of the molecule is attached to the primary terminal OH-group of sphingosine. This consists of a mucopolysaccharide residue, with building blocks of glucose, galactose, *N*-acetylgalactosamine and *N*-acetylneuraminic acid, in varying chain length and containing varying amounts of *N*-acetylneuraminic acid.

Based on the work of KLENK[3] and BOGOCH[4] and on his own investigations, SVENNERHOOM[5] assigned the structure of a tetrasaccharide (Fig. 1, I) to the neuraminic acid-free mucopolysaccharide residue of the major normal ganglioside, and a trisaccharide configuration (Fig. 1, II) to the corresponding residue of the Tay-Sachs' ganglioside. He established the chemical relationship between the ceramide tetrasaccharide and the ceramide trisaccharide and suggested that the ganglioside which accumulates in Tay-Sachs' disease cannot undergo normal metabolic breakdown (Fig. 1; Block B').

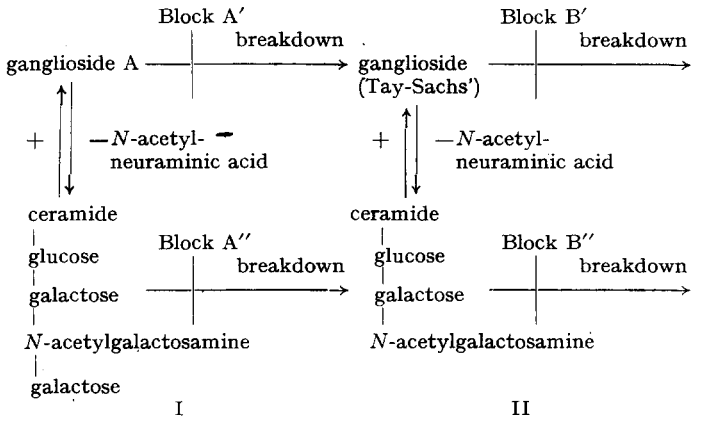

Fig. 1. Scheme of metabolic blocks in cases of infantile amaurotic idiocy. Block B' or B'': block giving rise to the storage of Tay-Sachs' ganglioside and Substance II. Block A' or A'': special form giving rise to the storage of ganglioside A and Substance I.

We have found a case of a biochemically special form of late infantile amaurotic idiocy which has been histologically verified[6] and is described below. In the brain

(preserved in formalin for 26 years) of the child Kn. the normal major ganglioside A* was stored, while only traces of Tay-Sachs' ganglioside could be detected, and Substance II (Fig. 1) was not found at all.

The storage of the Tay-Sachs' ganglioside is accompanied by an accumulation of the neuraminic acid-free ceramide trisaccharide (II) This Substance II has already been observed by other investigators[2,7] in these pathological brains; however, the exact relationship to the Tay-Sachs' ganglioside was never recognized. We have found

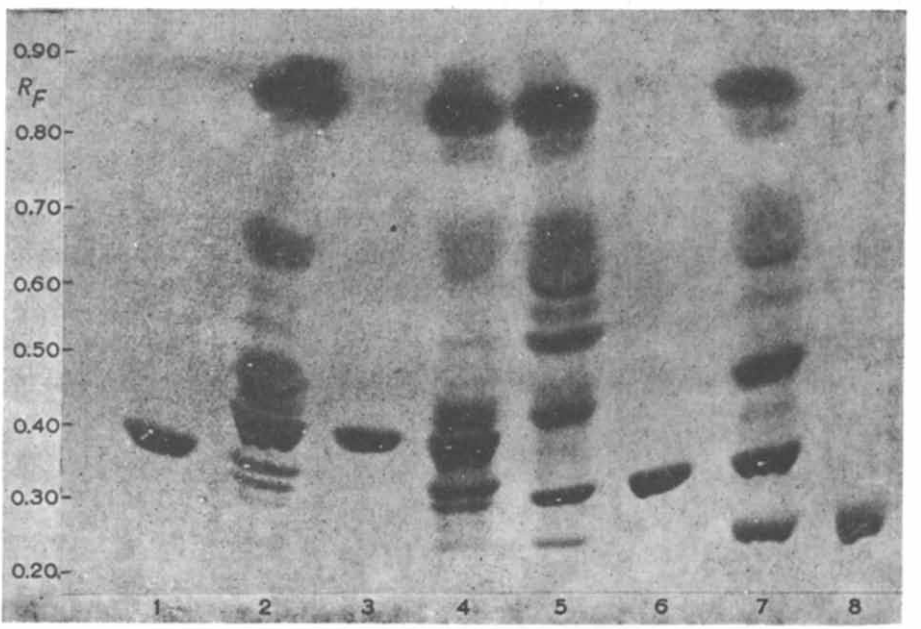

Fig. 2. Thin-layer chromatogram of lipid extracts of brain tissue from two usual infantile cases and from one special form of late infantile, amaurotic idiocy. Adsorbent: 400-μ-thick layer of Kieselgel G, Merck; solvent system: propanol – conc. ammonia – water (6:2:1); height of the solvent front: 15 cm; detection: anisaldehyde sulphuric acid in acetic acid (reagent of Kägi–Miescher). 1, 20 μg of neuraminic acid-free residue of ganglioside Tay-Sachs (R_F 0.33) (Fig. 1, II); 2, 250 μg of total lipid extract of the brain cortex in a case of infantile amaurotic idiocy (fresh tissue); 3, 20 μg of ganglioside Tay-Sachs (R_F 0.37); 4, 250 μg of total lipid extract of the brain cortex in a case of infantile amaurotic idiocy, preserved in formalin for 26 years; 5, 250 μg of total lipid extract of normal brain cortex, preserved in formalin for 26 years; 6, 20 μg of ganglioside A (R_F 0.30); 7, 250 μg of total lipid extract of the brain cortex in a special form of late infantile amaurotic idiocy, preserved in formalin for 26 years; 8, 20 μg of neuraminic acid-free residue of ganglioside A (R_F 0.26) (Fig. 1, I).

that the storage of ganglioside A is accompanied by a corresponding accumulation of the neuraminic acid-free ceramide tetrasaccharide (I). This also occurs as a minor component in normal brain tissue. From analysis of fresh brain tissue (Fig. 2), it seems unlikely that these substances have arisen from the gangliosides as a result of the long storage in formalin[8].

All our findings are consistent with the scheme, shown in Fig. 1. If it is assumed that an enzymic block occurs in the metabolic breakdown in infantile amaurotic

* Named by KLENK: ganglioside A, by KUHN: ganglioside G_2 and ganglioside G_3, and by SVENNERHOLM: major monosialoganglioside.

idiocy this block could exist either in the degrading of the gangliosides (Blocks A' and B') or in their corresponding neuraminic acid-free residues (Blocks A'' and B'').

The chemical characterization of the special form of amaurotic idiocy was performed by means of thin-layer chromatography (Fig. 2) and by comparison of the R_F values of the different substances in 4 solvent systems: chloroform – methanol – water (11:9:3); *n*-butanol – pyridine – water (6:4:3); 76 % phenol; and that indicated indicated in Fig. 2. The spray used for detection (Fig. 2), stained the different classes of lipids sensitively and specifically.

The gangliosides were isolated by KLENK's procedure[3], the appropriate neuraminic acid-free ceramide saccharides were obtained by self-decomposition[4,5] of the gangliosides in the acid form; they were isolated in a pure state by column chromatography on wet Florisil[9].

Biochemische Abteilung der
Deutschen Forschungsanstalt für Psychiatrie,
Max-Planck-Institut, München (Germany)

HORST JATZKEWITZ
KONRAD SANDHOFF

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