

GENERALIZED GANGLIOSIDOSIS IN MALTA

E. A. CACHIA,

M.B., Ch.B. (Manch.), D.C.H.

J. MIFSUD,

M.D., D.Bact., M.R.C.Path

L. VASSALLO,

B.Sc., M.D., F.R.C.P. (Ed.), M.R.C.P. (Lond.), M.R.C.P. (Glasg.)

P. VASSALLO AGIUS,

M.D., D.C.H., M.R.C.P.

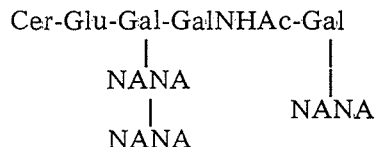
St. Luke's Hospital

Tay, a British ophthalmologist, described a cherry-red spot in the fundus of a retarded infant in 1881 and Sachs a few years later established the basic clinical picture of the syndrome called amaurotic family idiocy.

The abnormal deposits of glycolipids in neurones termed ganglioside by Klenk in 1940, appeared to be the basic pathologic abnormality responsible for the disease. Visceral lipidosis was not a feature of the classical disease described by Tay and Sachs. However, atypical cases of Tay Sachs and others resembling Niemann Pick were also described. The right solution for these problems has been recently provided by the neurochemist.

The key ganglioside has the following chemical structure: *

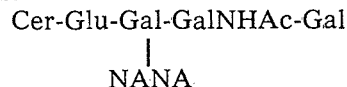
* Cer = ceramide; Glu = glucose; Gal = galactose; GalNHAc = N-acetyl galactosamine; NANA = N-acetylneuraminic acid (= sialic acid)



The metabolism of this substance initially involves the removal of one or other molecule of sialic acid by specific lysosomal hydrolytase enzymes. Further down the metabolic pathway the terminal galactose residue is removed, again by a specific enzyme.

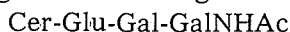
At least nine different main gangliosides can normally be detected in brain tissue and various diseases have been attributed to the abnormal accumulation of a ganglioside or a related compound, due to absence or deficiency of the specific enzyme.

The chemical structure of ganglioside GM₁ is:



There are two types of GM₁ gangliosidosis. Type 1 (generalized gangliosidosis) is characterized by deficiency of the degradative enzymes B-galactosidase A, B, and C; and Type 2 (late onset GM₁ gangliosidosis) due to absence of B-galactosidase B and C.

GM₂ ganglioside, which is the next step in the metabolic pathway, is very closely related, differing from GM₁ by lacking the terminal galactose residue:



Thus in Tay-Sachs disease (GM₂ gangliosidosis, Type 2, there is absence of galactosaminidase A (or hexosaminidase A) with cumulation of GM₂. In GM₂ gangliosidosis, Type 2, there is absence of hexosaminidase A and B. Type 3 is a late onset type characterized by partial deficiency of hexosaminidase A.

By losing GalNHAc, GM₂ becomes GM₃, viz:



These three gangliosides GM₁, GM₂ and GM₃, on losing their sialic acid group give rise to other gangliosides termed GA₁, GA₂ and GA₃ respectively (Raine, 1969).

The purpose of this paper is to describe the clinical features of four cases of generalized gangliosidosis, two of which in siblings. In two of the cases, and in one of the siblings, B-galactosidase deficiency was demonstrated in blood and urine samples.

Case Reports

Case I. — D.C.I. Male — Born 28. 11. 69

This was the third child of unrelated young Maltese parents. The first child was normal and the second had been mentally retarded and had died aged 13 months (see Case 2).

He was a full term spontaneous normal delivery following a normal pregnancy. B.W. 8 lb. 8 oz. He was noted to have a high-pitched cry soon after birth and marked oedema of face, upper and lower

limbs, and scrotum. The oedema extended up to the elbows in the upper limbs, and up to the thighs in the lower limbs. There was no oedema of the abdomen. There was a narrow chest with anterior bowing of the sternum. His general condition was otherwise good and there were no feeding problems. Blood film examination at 3 days of age showed cytoplasmic vacuolations in about 60% of the lymphocytes. Though the liver and spleen were no more than normally palpable at birth, there was progressive enlargement of these organs within the next few months. Examination of the fundi showed cherry-red spots in the macular regions. He smiled at 3 months and laughed at 4 months. At 5 months he was fully investigated at the Hospital for Sick Children at Great Ormond Street, where B-galactosidase deficiency was diagnosed; α -glucosidase and hexosaminidase levels were normal.

His general condition slowly deteriorated and he died aged 13 months. He never had fits.

No P.M. was carried out.

Case 2. — N.C.I. — Born May 1968

This was the elder sister of Case No. 1. She was delivered normally at term following an uneventful pregnancy. There were no neonatal problems. Her development was apparently normal until about 6 months of age when she was noted to be hypotonic and mentally retarded, and to have hepatomegaly. Routine blood film examination at this time showed cytoplasmic vacuolations of lymphocytes, and a presumptive diagnosis of Tay-Sachs' disease with visceral involvement was made.

She developed convulsions at 11 months of age for which she was admitted to hospital. Clinical assessment at this stage (May 1969) showed an irritable baby with a vacant stare and she appeared blind. The neck muscles were hypotonic and she could not hold up her head. The anterior fontanelle was normal and rotatory nystagmus was present. The fundi showed greyish abnormal discoloration of the macular areas. There was no papilloedema. There were choreiform movements of the upper limbs while the lower limbs

were held motionless in abduction. Wasting of the thigh muscles was evident. The tendon reflexes in upper and lower limbs were normal. Plantar responses were flexor. The liver was enlarged 2 fingers below the right costal margin but the spleen was not enlarged. The lymph nodes were not palpable. The heart and lungs were clinically normal.

She continued to deteriorate and died one month later of pneumonia.

Post-mortem examination

She was a well developed female consistent with her age. The heart was moderately enlarged, with dilatation of both ventricles. The liver was moderately enlarged, with a slight yellowish discoloration on section. The spleen, thymus, lymph nodes and kidneys were macroscopically normal.

The cerebral hemispheres were oedematous and showed deep wide prominent sulci with venous congestion. There was marked ventricular dilatation. The lungs showed right upper lobe consolidation.

Histology: Abnormal foam-cells were seen in the kidneys, liver, spleen and lungs.

Case 3 — R.M. Female — Born 22. 11. 71

This was the second child of unrelated parents. The first child is 2½ years old and suffers from bronchial asthma. She was normally delivered at term at home after a normal pregnancy. She cried well after delivery and was noted to have bilateral wrist drop. The baby was otherwise normal. The wrist-drop improved with splints and physiotherapy. The baby progressed normally until 10 weeks old when she was admitted to St. Luke's Hospital with a painful right thigh accompanied by a rise in temperature. There was no enlargement of the liver and spleen at this time. A clinical diagnosis of osteomyelitis was made. There was no evidence of this on radiological examination. Blood film at this time showed the cytoplasmic vacuolations in the majority of the lymphocytes. Examination of the fundi revealed no cherry-red spots. Blood and urine from patient showed absence

of B-galactosidase activity; diminished activity of this enzyme in the blood was reported in both parents and in the patient's brothers.

Her general condition continued to deteriorate and she died aged 7 months. No fits were observed.

Permission for post-mortem examination was not given.

Case 4 — M.C. Female

This was the second child of unrelated parents. She had been delivered normally after a full term normal pregnancy. There was no difficulty in onset of respiration and no abnormality was discovered in the neonatal period. She was subsequently difficult with feeds and at 4 months of age was still unable to hold her head up straight. On examination at this stage she was found to be obviously mentally retarded with generalized hypotonia and hepatosplenomegaly. She also had diffuse slate-coloured areas of pigmentation, similar to "mongolian blue spots", at the back of the trunk, less marked on the abdomen. Examination of the blood film showed cytoplasmic vacuolations in a high proportion of the lymphocytes. Blood examination showed normal hexosaminidase level but very small activity of B-galactosidase was present; this latter enzyme was not found in the urine. The parents had normal levels of these enzymes.

The child was admitted to hospital at the age of 9 months in status epilepticus. Her condition remained poor and she had numerous further convulsions until her death 7 months later with a chest infection.

Post-mortem examination was refused.

In summary, the clinical features were as follows (*Table*). All cases were the products of unrelated Maltese parents. Two of the cases were brother and sister. No abnormality of pregnancy has been noted and the deliveries have been uncomplicated. Two cases were ostensibly normal at birth, but one had generalized oedema, more marked in the lower limbs, hands and eyelids; another case had bilateral wrist drop at birth. The only male of the four cases had gross bilateral hydro-

coele. One case had painful right thigh at two months of age. Mental development was definitely retarded by six months of age in all cases. Enlargement of the liver was a feature in all cases. Fits are a late feature of the disease: three cases had fits by nine months of age; the one who did not have fits died at seven months. Examination of the fundi showed cherry-red spots or a greyish band in the region of the macula in three cases. In all instances cytoplasmic vacuolations of the lymphocytes were present in the peripheral blood; in one case, who had a sibling affected, these were seen at 3 days of age.

TABLE

Summary of Cases 1-4

Common Features

- Parents unrelated.
- Two of the cases were siblings.
- Normal pregnancy, delivery and birth weight.
- Mental retardation.
- Hepatomegaly.
- Cytoplasmic vacuolations in lymphocytes.
- Early death.

Other Features

- | | |
|--|-----|
| Fits. | 2/4 |
| Abnormal fundi. | 3/4 |
| Oedema in neonatal period. | 1/4 |
| 'Extensive M.B. spots'. | 1/4 |
| Gross bilateral hydrocoele: Present in only male case. | |

We are also aware of six other cases with similar clinical features and cytoplasmic vacuolations in the lymphocytes. However, these are not included here because no enzyme studies could be carried out.

Discussion

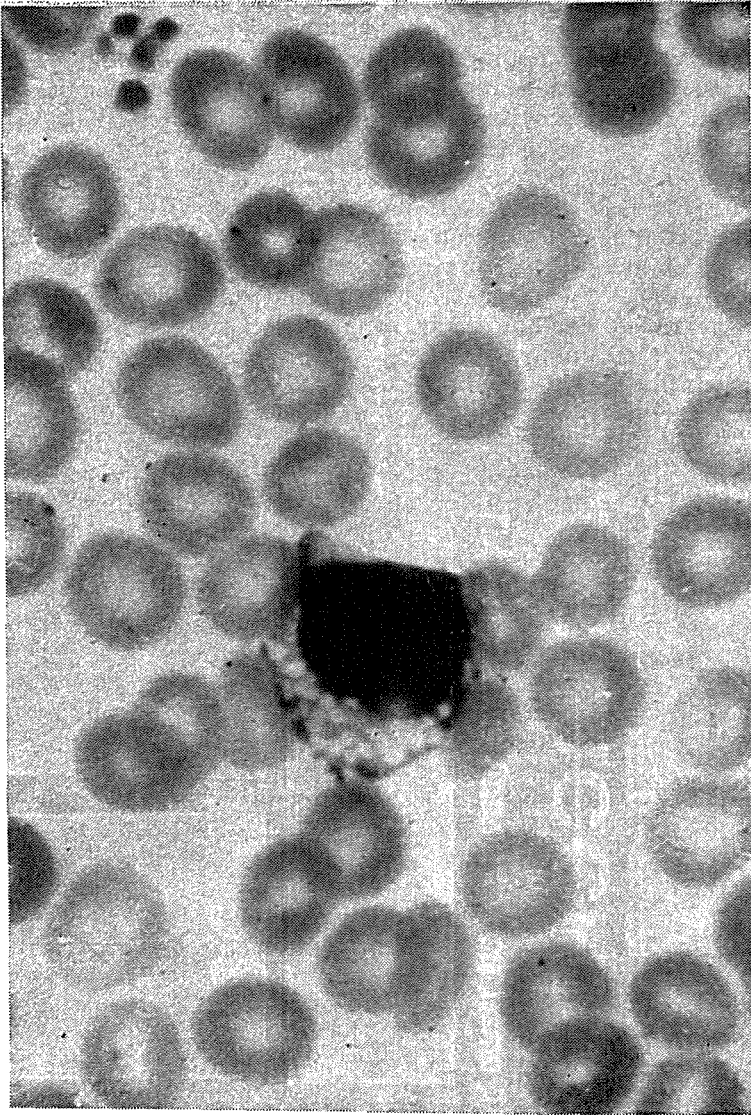
This condition is a metabolic disorder, one of the lipidoses, transmitted as an autosomal recessive. In affected children, supposedly homozygous with respect to the abnormal gene, a high proportion of the lymphocytes in the peripheral blood contain numerous prominent, typical cyto-

plasmic vacuoles (*See figure*). Such vacuolations may, however, also be noted, to a much lesser degree, in monocytes and, even more rarely, in polymorphonuclear neutrophils and eosinophils. These vacuoles are typically seen packing the cytoplasm of the lymphocyte concerned; they appear perfectly round with sharply demarcated outlines; this distinguishes them from the 'degenerative' vacuolations which are fairly frequently noted in mononuclear cells in peripheral blood films.

Rayner and Boök (1958) examined the blood of 29 children having this disease and found all to have these abnormal cells. They also showed that parents of affected children frequently have a few of these lymphocytes in their blood. Being themselves normal and supposedly heterozygous for the abnormal gene, the parents presumably have fewer abnormal cells.

The nature of the vacuoles is not known since they are not stainable by any histochemical method. Ganglioside accumulates in the brain in abnormal amounts in these diseases and it has been suggested therefore that the vacuoles in the lymphocytes may also contain this glycolipid material.

The point that emerges from these cases is the realization that the clinical distinction between the various ganglioside lipidoses is now no longer possible, even in the clinically 'typical' case — the resemblance to one or other of the classical Tay-Sachs or Niemann Pick is meaningless without the aid of specialised enzymatic tests. A diagnosis of Tay-Sachs disease with visceral involvement is nowadays obsolete and incorrect. Moreover, the dramatic finding of the cherry-red spots in the retina is now no longer pathognomonic of classical Tay-Sachs disease, as was formerly widely held by clinicians. The cherry-red spot is now regarded only as a cherry-red herring as far as an exact diagnosis is concerned. The only reliable differential diagnostic tool which has assumed a *sine qua non* status is that of specialised enzymatic studies for the specific deficiencies, as in our cases. The presence of vacuolations



Peripheral blood film showing typical cytoplasmic vacuoles.

in the lymphocytes is a useful screening test and should alert the haematologist and, through him, the clinician as to their diagnostic significance, necessitating further special enzymatic studies. These vacuolations have, in one of our cases, been present in the neonatal period. Indeed, in a family with a previously affected child, the presence of these vacuolations in the peripheral blood of the neonate is sufficient evidence that the child is going to be affected by this fatal condition.

We would also like to emphasise that equally well one cannot rely on histolo-

gical appearances to establish the exact nature of a lipidosi such as Tay-Sachs or Niemann Pick, even if the appearances are characteristic.

Conclusion

Many of the cases which used to be grouped together as 'amaurotic family idiocy' now emerge as different metabolic defects, however similar or identical the clinical picture might be. Therefore in every case the exact metabolic defect should be pin-pointed before a definite diagnosis can be made. It therefore

becomes mandatory to investigate all patients in this group by special chemical techniques. The presence of cytoplasmic vacuolations in the lymphocytes, with the characteristics described in this paper, should be an indication for performing further special enzyme studies.

Addendum

Since writing this paper we have had two more cases of generalised gangliosidosis proved by specific enzyme studies. One case presented with facial and distal limb oedema at birth; the other with mental retardation, generalised hypotonia and hepatomegaly at eight months of age. In both cases the characteristic cytoplasmic

vacuolations were seen in the lymphocytes in a peripheral blood smear.

Acknowledgement

We thank Professor B. Clayton and her staff at the Institute of Child Health, London, for carrying out the enzyme studies.

References

- KLENK, E. (1940) *Z. Physiol. Chem.* **262**, 128
 RAINE, D.N. (1969) *Lancet*, ii, 959
 RAYNER, S., and BOOK, J.A. (1958) *Lancet*, i, 1077
 SACHS, B. (1887) *J. Nerv. and Ment. Dis.* **14**, 541
 SVENNERHOLME, L. (1963) Chromatographic separation of human brain gangliosides, *J. Neurochem.* **10**, 613
 TAY, W. (1881) *Tr. Ophth. Soc. U. Kingdom* **1**, 155