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Clinical and neuropathological picture of ethylmalonic aciduria – diagnostic dilemma

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Abstract

Increased ethylmalonic acid (EMA) in urine is a non-specific finding, and is observed in a number of inborn errors of metabolism, as well as in individuals who carry one of two common polymorphisms identified in the SCAD coding region. The authors present an 8-month-old girl with a suspicion of neuroinfection, although the clinical presentation led to diagnosis of ethylmalonic aciduria. From the neuropathological point of view the most remarkable changes were observed in the brain cortex, which was diffusely damaged practically in all regions of the brain. Of note, the most severe destruction was observed in the deepest regions of the sulci. The cortex of the affected regions showed no normal stratification and its structure was almost totally replaced by a form of "granulation tissue" with a markedly increased number of capillaries. To the authors' knowledge this is the first clinical report of ethylmalonic aciduria with brain autopsy findings.

Key words: ethylmalonic acid, ethylmalonic aciduria, clinical report.

Introduction

Increased ethylmalonic acid in urine (EMA) is a non-specific finding, and is observed in a number of inborn errors of metabolism, as well as in individuals who carry one of two common polymorphisms identified in the SCAD coding region [2,8]. Ethylmalonic aciduria may be seen in short-chain acyl-coenzyme A dehydrogenase (SCAD) deficiency. It is also observed in patients with other disorders of mitochondrial fatty acid oxidation, including the ethylmalonic encephalopathy syndrome, glutaric acidaemia type II (deficiency of electron transfer flavoprotein and of electron transport flavoprotein – ubiquinone oxidoreductase), and a large number of still biochemically uncharacterized patients with persistent elevated urinary EMA.

Ethylmalonic encephalopathy (gene map locus: 19p13.32, mutations in the ETHE1 gene; MIM #608451) coexists with MRI changes including

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increased signal on T2-weighted images in the putamen, caudate nuclei and periaqueductal region, which correspond to symmetrical necrotic lesions [1,5,12,20]. The patients occasionally have signal abnormalities in subcortical white matter areas, and brainstem, cerebral atrophy in frontal and temporal regions, and cerebellar atrophy [9,12,14]. In some children tethered cord, hygroma/haematoma and a Chiari type I malformation may be found. Additional biochemical abnormalities include elevated plasma concentration of C4 and C5, acylcarnitine and sometimes elevated lactic acid with cytochrome oxidase deficiency in muscle biopsy. The most frequent clinical features observed in ethylmalonic encephalopathy are petechiae, orthostatic acrocyanosis and chronic diarrhoea [3,8]. Neurological assessment usually shows developmental delay, microcephaly, cerebellar ataxia, in the beginning hypotonia then pyramidal and extrapyramidal dysfunction [6,16].

The literature data lead to the assumption that ethylmalonic aciduria is a very rare disorder. However, the actual incidence of the condition may be significantly underestimated because the biochemical phenotype may be incorrectly attributed to other metabolic disorders [15,18].

Patient presentation

An 8-month-old girl, the first daughter of young, unrelated parents of Polish origin. The gestation period was complicated by arterial hypertension in the mother. The girl was born by vaginal, uneventful delivery (birth weight of 3130 g, Apgar score of 10 points). In the family history febrile seizures were observed. The psychomotor development was normal since the 6th month of life. At the age of 6 months a sudden episode of hypotonia and loss of reaction to external stimuli in the course of viral infection was noticed (contact with chickenpox in anamnesis). A month later during bronchopneumonia and temperature of 39°C status epilepticus was observed. Due to cardio-respiratory insufficiency the girl was admitted to the intensive care unit. Based on suspicion of neuroinfection antibiotics and acyclovir were introduced. In treatment mechanical ventilation, catecholamines, thiopental, phenytoin, phenobarbital and valproic acid were used. CT of the head showed mild cerebral atrophy in the frontal lobes. The cerebrospinal fluid examination performed three times was normal. PCR Herpes simplex was normal in plasma and cerebrospinal fluid. Adenovirus infection was also excluded.

Because of suspected neuroinfection after 4 days the child was transferred to the Child Neurology Department. At admission the girl was sleepy, without visual contact, and with weight deficiency and hepatomegaly. Neurological examination showed head circumference of 44 cm, frontal fontanelle 4x4 cm, at the level of the cranial bone, spastic tetraparesis, without clinical signs of increased intracranial pressure. MRI showed contrast enhancement hyperintensity of the globus pallidus, parietal and occipital lobes, less marked in the frontal lobes and cerebellum [Figs. 1-8]. The changes observed in MR imaging implied the suspicion of neuroinfection (Herpes simplex?). The repeated MRI (after 2 weeks of treatment) showed a small progression of changes especially conspicuous in the temporal regions [Figs. 1-8]. Immunoglobulins were added to the therapy regimen. In EEG generalized paroxysmal changes were evident. Polymorphic seizures (tonic seizures, partial secondary generalized seizures) were treated with phenytoin, clonazepam, and valproic acid. In laboratory examinations increased transaminases (GPT 134 U/l, GOT 317 U/l), lactic acid (4.95 mmol/l), lactate dehydrogenase (1824 U/l), and thrombocytosis (1223 K/uL) were seen. Organic acid analysis in urine revealed increased excretion of ethylmalonic acid and slightly increased adipic acid. Increased plasma butyrylcarnitine (C4) was proved by tandem mass spectrometry. On the 30th day of hospitalization the child experienced high fever, recurrent vomiting, seizures, oedema, and oliguria, followed by acute cardio-respiratory failure. After resuscitation the girl was transferred to the ICU, where after several hours she died. An autopsy was performed with special attention to the central nervous system.

From the neuropathological point of view the most remarkable changes were observed in the brain cortex, which was diffusely damaged practically in all regions of the brain. Of note, the most severe destruction was observed in the deepest regions of the sulci [Fig. 9]. The cortex of the affected regions showed no normal stratification and its structure was almost totally replaced by a form of "granulation tissue" with a markedly increased number of capillaries. Both in the white matter and in the cortex marked astrogliosis was seen [Fig. 10]. Interestingly, in the most severely affected regions, gliosis was almost absent

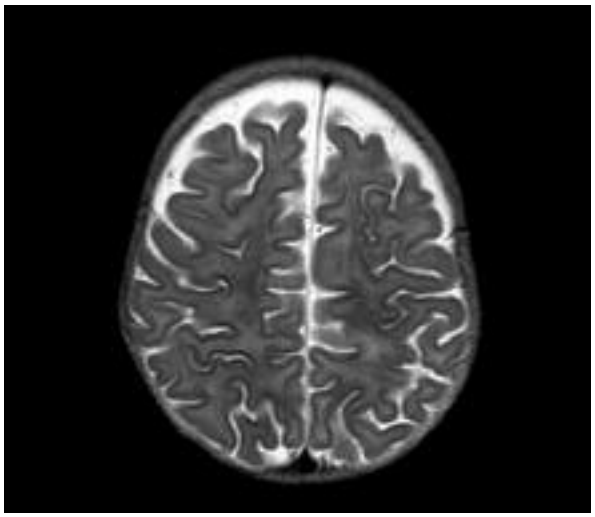

Fig. 1. SE/T2 weighted MR images – lack of myelination pattern in white matter of frontal lobes, thin cerebral cortex.

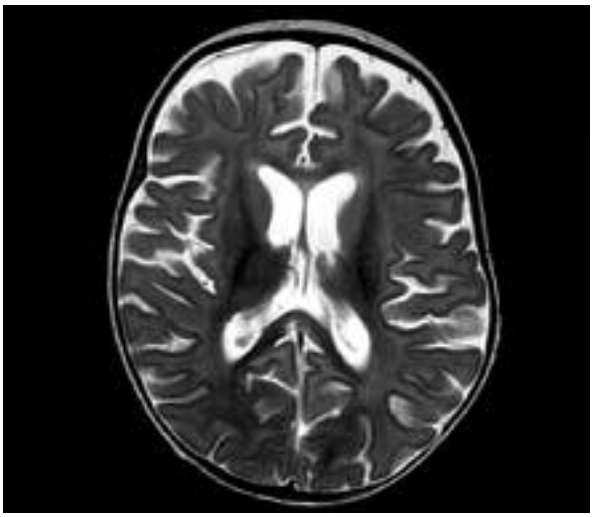

Fig. 2. SE/T2 weighted MR images – myelination pattern seen in internal capsules and partially in corpus callosum. The remaining brain structures do not show a proper myelination process. Increased cerebral sulci with increased subarachnoid spaces in frontal areas.

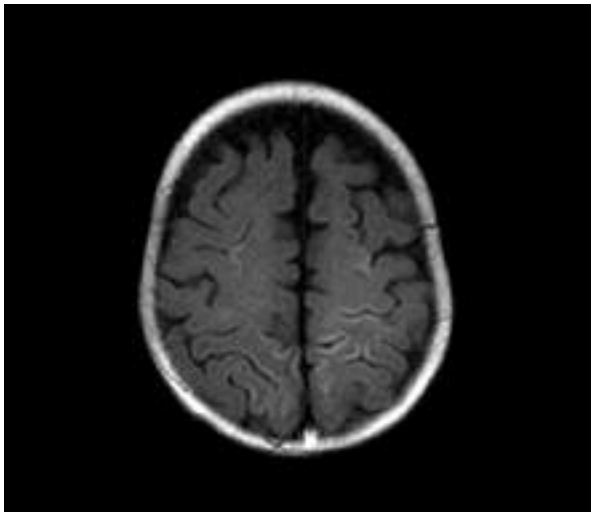

Fig. 3. SE/T1 MR weighted images – hyperintense areas visible in frontal cortex indicative of cortex passive hyperaemia.

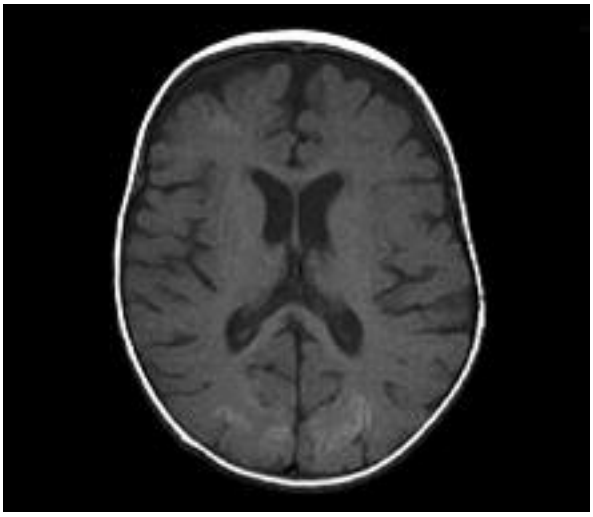

Fig. 4. SE/T1 weighted MR images – high intensity of signal areas seen in cerebral cortex of right frontal and both occipital lobes indicative of passive hyperaemia of the cortex.

(no GFAP-positive astrocytes), which strongly contrasted with remarkable gliosis in the vicinity of these areas of total destruction of the cortex. In fully developed necrotic areas numerous macrophages were observed in the cortex as well as in the white matter, occasionally forming dense aggregations. In the whole brain including the cerebellum there was marked hypomyelination with gliosis, especially evident in GFAP immunohistochemistry.

The cerebellum was also evidently affected with apparent hypoplasia (or atrophy) [Fig. 11]. Purkinje cells were only sporadic and Bergman gliosis was conspicuous. The internal granular layer was paucicellular. Focally remnants of fetal external granular

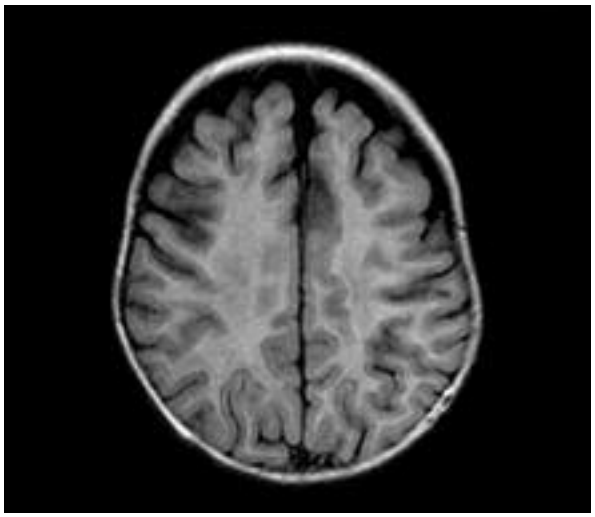

Fig. 5. FLAIR weighted MR images – white matter intensity of signal does not show pathological changes, thin cerebral cortex in frontal areas.

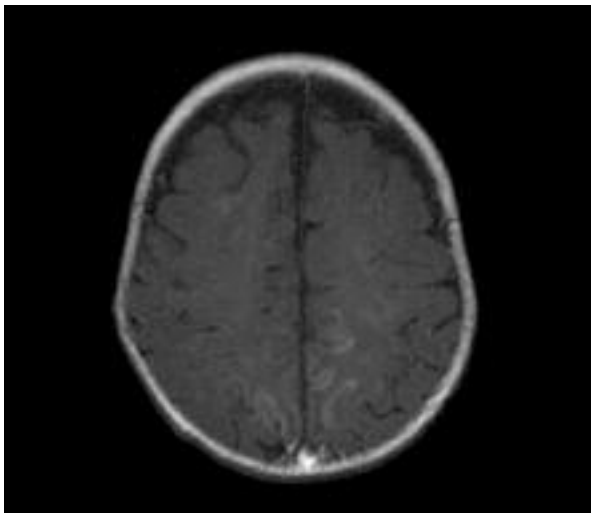

Fig. 6. SE/T1 weighted MR images after enhancement – in frontal cortex hyperintense signals (more visible after contrast enhancement) suggestive of passive hyperaemia of the cortex.

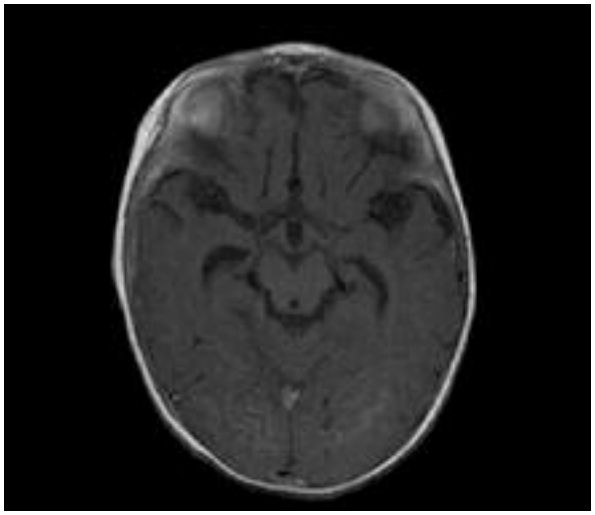

Fig. 7. SE/T1 weighted MR images after enhancement – in the cortex of occipital lobes more visible hyperintensity of signal suggestive of cortex passive hyperaemia.

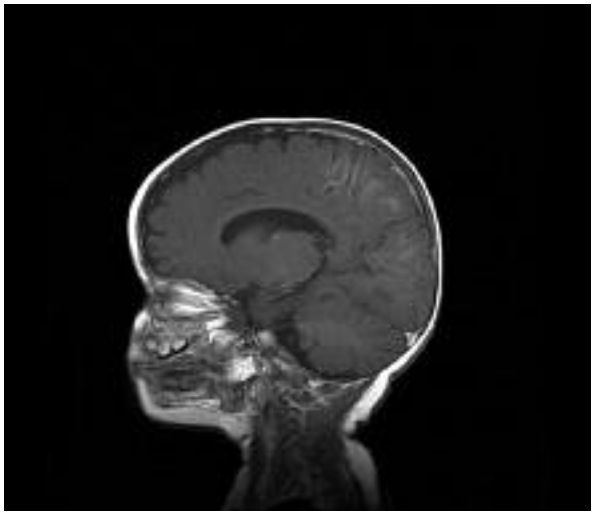

Fig. 8. SE/T1 weighted MR image after enhancement – hyperintense linear areas in the cortex of parietal and occipital lobes (right side) suggestive of cortex passive hyperaemia.

layer were observed. Some hypercellularity was noted in the molecular layer of the cerebellar cortex, which probably reflected a sort of retarded (or arrested) migration from the external granular layer [Fig. 12]. Deep nuclei of the cerebellum and the brain stem showed some neuron loss with concomitant gliosis. In comparison to the brain cortex, basal ganglia were relatively much less affected, with only a little hypomyelination and loss of neurons.

In the general autopsy the liver was the most affected organ, showing signs of extensive steatosis of hepatocytes [Fig. 13] and scanty lymphocytic periportal infiltrations (not shown).

Discussion

Ethylmalonic aciduria has been described in single case reports. To the authors' knowledge this is the

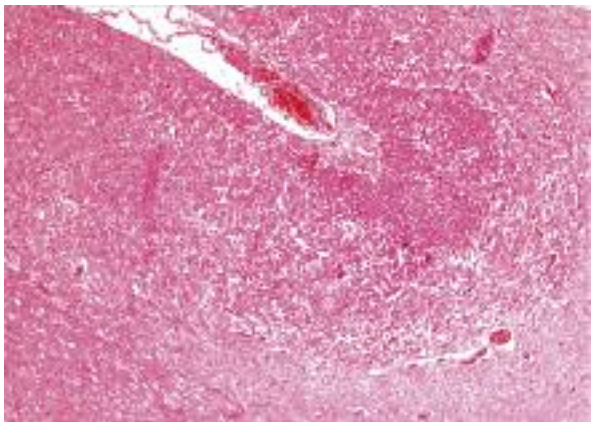

Fig. 9. Cerebral cortex most severely damaged in the region of the bottom of the sulcus, with somewhat partially preserved lateral parts of sulcus. Haematoxylin-eosin.

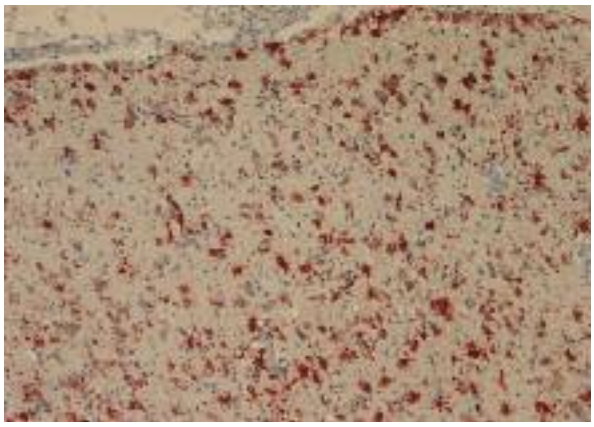

Fig. 10. Massive astrogliosis in cerebral cortex. GFAP immunohistochemistry.

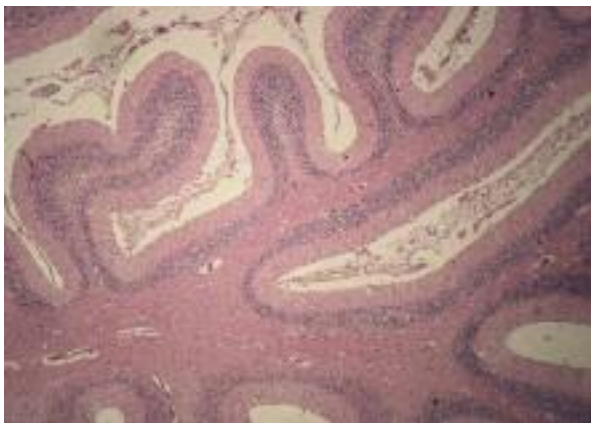

Fig. 11. Cerebellum with features of conspicuous hypoplasia. Haematoxylin-eosin.

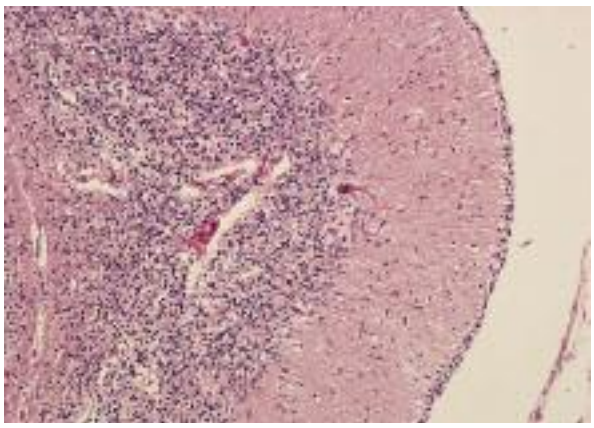

Fig. 12. Cerebellar cortex. Remnants of external granular layer, moderate hypercellularity in molecular layer and marked loss of Purkinje cells are noted. Haematoxylin-eosin.

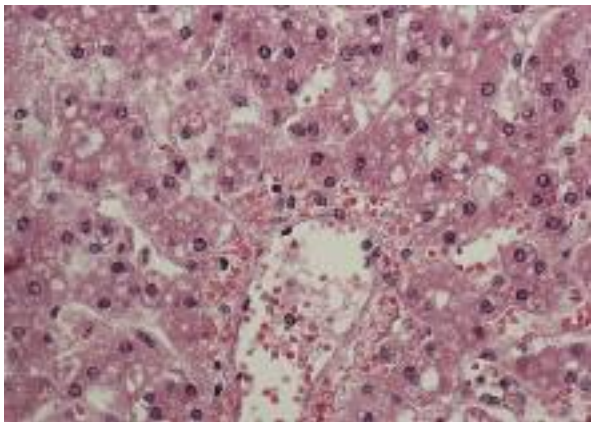

Fig. 13. Microdroplet steatosis of hepatocytes. Haematoxylin-eosin.

first clinical report with brain autopsy findings. Ethylmalonic aciduria is most often seen in SCAD deficiency and ethylmalonic encephalopathy. Di Rocco *et al.* suggested that ethylmalonic aciduria is not a constant biochemical marker of ethylmalonic encephalopathy and that its normal excretion outside of metabolic decompensation episodes does not exclude this metabolic disease [13,16]. Biochemically, all patients have shown elevated excretion of ethylmalonic acid (EMA) in urine, supposedly from carboxylation of accumulated butyryl-CoA.

Over 20 confirmed cases of short-chain acyl-CoA dehydrogenase (SCAD; McKusick 606885) deficiency

(McKusick 201470) have been published, demonstrating an extraordinarily wide range of clinical symptoms [16,20]. One group of patients showed mainly episodes of acute, recurrent metabolic crises including hypoglycaemia, metabolic acidosis, vomiting and drowsiness [1,4,17,22]. A second group predominantly demonstrated a neuromuscular picture with myopathy and hypotonia, and seizures with or without mental retardation [2,3,4,6,7]. In some cases a sort of temporary swinging of symptoms typical for the first and second group was observed [10,11]. A patient with muscle weakness combined with ophthalmoplegia and multicore myopathy has been described [19,20].

In our patient electron transfer flavoprotein and electron transport flavoprotein dehydrogenase deficiencies were excluded by absence of glutaric and isovaleric acids in the urine with isovaleryl and hexanoyl glycines. Due to quick progression of symptoms the presented patient was not subjected to either enzymatic or molecular study.

In neuropathological examination the cortical changes cannot be regarded as specific. These changes could be of ischaemic nature (the child was reanimated twice) but the diffuse character and not particularly specific topography of cortical damage as well as its severity with enormous glial reaction suggest that general metabolic derangements played an important role in the brain pathology.

The morphological picture should be differentiated from Alpers-Huttenlocher syndrome. However, although no changes were found that could indicate an inflammatory process (as in Alpers syndrome), firstly, mild liver changes limited to fatty changes as in our case occur in only a minority of cases of Alpers syndrome (in those children the picture of nodular cirrhosis of the liver is typical), secondly, in Alpers syndrome loss of neurons, extensive vacuolation of the cortex and astrocytosis are the dominant neuropathological changes, and thirdly, as already mentioned, the severity and distribution of changes (with relatively spared gyral crests) indicate the ischaemic/metabolic nature of the pathology in the reported case [10,20].

Increased concentration of ethylmalonate, along with methylsuccinate, may be the only urinary organic abnormalities in patients with SCAD deficiency. Ethylmalonic aciduria without methylsuccinate is also found in oxidase cytochrome c deficiency. The combination of lactic acidosis, ethylmalonic and methylsuccinic aciduria along with the excretion of butyrylglycine, isovalerylglycine and 2-methylbutyrylglycine are characteristic for ethylmalonic aciduria. Despite the suspicion of neuroinfection, the clinical presentation, laboratory tests and autopsy results most likely point to the diagnosis of SCAD.

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