Precision medicine in ethylmalonic encephalopathy: patient derived fibroblasts for physiopathological research and finding potential therapies

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INTRODUCTION AND PURPOSE

Precision medicine fucuses on using cells directly from patients to characterize their phyisiopathology and try to find out potential therapies.

Ethylmalonic encephalopathy (EE) is a serious metabolic disorder that usually appears in early childhood development and primarily effects are seen in the brain, gastrointestinal tract, and peripheral vessels. This disease is usually progressive and fatal. EE is characterized by high levels of acidic compounds in body fluids. Another feature of the disease is that the activity of mitochonddrial complex IV is usually decreased, which limits energy production in tissues that require a large energy supply such as neuronal and muscle tissues.

The main common point of all the metabolic alterations related to EE is that a mitochondrial sulfur dyoxigenase named ETHE1 is altered, what leads to accumulation of H2S due to its deficient elimination.

MATERIALS AND METHODS

In this work, fibroblasts of a patient that suffers from ethylmalonic encephalopathy are used in order to characterize the physiopathology. These cells are used in various experimental assays with the purpose of knowing the metabolic pathways that are affected, such as sulfur detoxification through ETHE1 and other alterations induced by the accumulation of H2S such as complex IV function and high concentrations of organic short-chain acids such as ethylmalonic acid. Some of these assays are Western blots in order to know the expression levels of mitochondrial proteins, respiratory complexes activity, intracellular sulfur concentration and, finally, pharmacological screenings with the goal of identifying positive compounds. We used a cocktail (CK1) to improve the physiopathology of the patient.

RESULTS

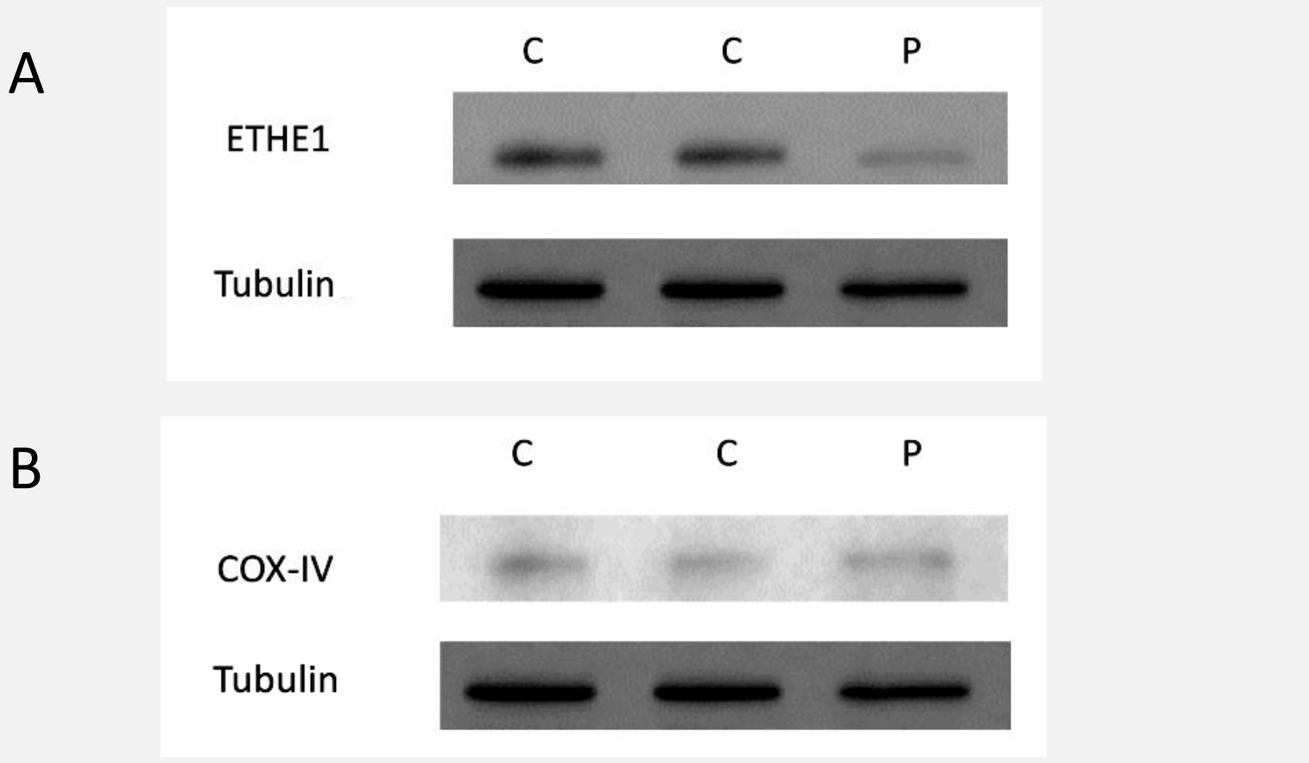

Fig. 1. Characterization of expression levels of proteins in the patient. A) Caracterization of ETHE1. **B)** Characterization of COX-IV. Tubulin was used as a charge control. (C: control, P: patient)

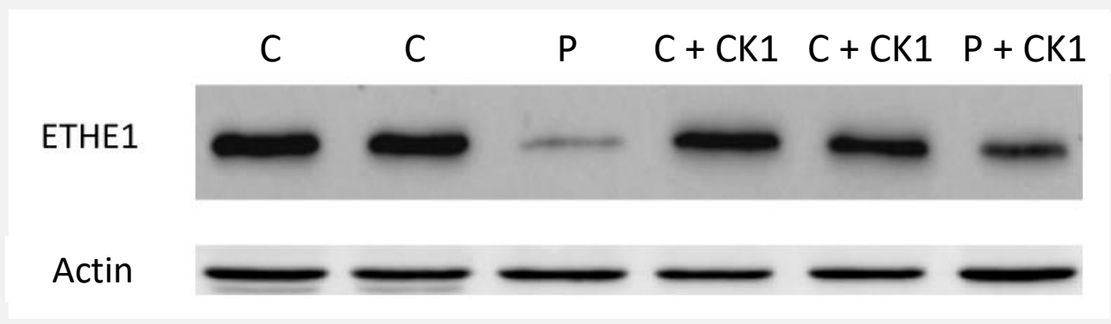

Fig. 2. ETHE1 expression with CK1 treatment. Western Blot analysis for comparing ETHE1 expression in the patient an in the patient with CK1 treatment. Actin was used as charge control. (C: control, P: patient, C + CK1: control with CK1 treatment, P + CK1: patient with CK1 treatment)

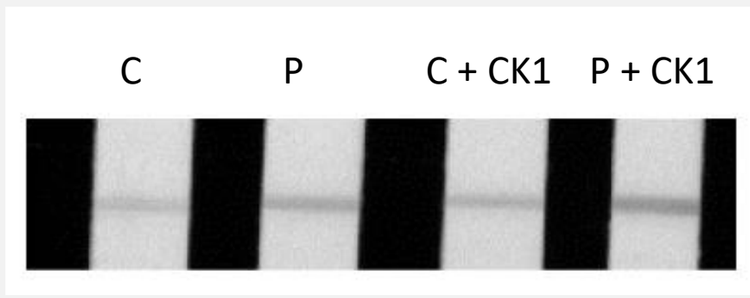

Fig. 3. Measurement of the activity of mitochondrial complex IV with and without CK1 treatment. The intensity of the bands is directly proportional to the activity of the mitochondrial complex IV. (C: control, P: patient, C + CK1: control with CK1 treatment, P + CK1: patient with CK1 treatment)

CONCLUSIONS

- Fibroblast cell cultures derived from patients are interesting cellular models for both disease modelling and pharmacological screenings.
- The expression of the enzyme ETHE1 is decreased in this patient.
- The activity of complex IV is not decreased in the patient.
- The treatment with CK1 improves ETHE1 expression and complex IV activity
- It is necessary further studies to confirm the results and to clarify the mechanism of action of the positive compounds and their use as medical therapies.

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