

Acetylation of HSP20 and lysine acetylases (KATs) in the human myometrium

Background

Preterm birth is childbirth that occurs before 37 weeks of pregnancy. Approximately 45–50% of preterm births are idiopathic, meaning that the cause is unknown.

• The muscle cells in the myometrium, which is the muscular wall of the uterus, is where research is conducted to understand the onset of labour.

The mechanisms that lead to early labour are poorly understood, and there is no effective treatment available.

The small heat shock protein 20 (HSP20) has been shown to be implicated in keeping the myometrium in a relaxed state, this occurs by phosphorylation of the protein. It is also known that this protein may be activated by acetylation and this may have a similar effect.

Introduction

Lysine acetylation has been thought to play mainly a role in regulating gene expression via histone acetylation and be only concentrated in the nucleus. On a closer look, there seems to be many lysine acetylation sites that also exist in the proteins located within the cytoplasm. Therefore the acetylation of lysine might have a larger role in regulating cell physiology (1). Therefore, acetylation of the cytoplasmic protein HSP20 may be implicated in the quiescent state of the myometrium.

The aim of this research was to investigate the levels of HSP20 and acetylated (ac) HSP20 proteins in myometrium obtained from pregnant (P) and nonpregnant (NP) women. Additionally the levels of CBP and KAT9, the lysine acetyltransferases (KATs) were investigated in this tissue.

Materials and methods

• Tissues were homogenized in WB buffer. 10-50ug of proteins were denatured in loading buffer at 95° C and separated on polyacrylamide gels. Proteins were then transferred onto a nitrocellulose membrane and incubated with respective antibodies.

Results

a)

b)

HSP20

Significant increase in expression of HSP20 was observed in P compared with NP tissues. HSP20 (Millipore, 1:20 000, 1h RT)

AcHSP20

No acHSP20 was observed in NP tissue, while high amounts were found in P tissue. acHSP20 4G (Abcam, 1:150 ON 4°C)

KAT 9

This KAT is expressed in all P samples and also in 4 out of 6 NP samples. KAT 9 (Abcam, 1:1000 ON 4°C)

d)

C)

CBP

This KAT is expressed in all P samples and is also expressed in 3 out of 6 NP Samples. CBP (Abcam, 1:3000 1h RT)

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• Tissue samples were collected from pregnant and non-pregnant human myometrium.





Discussion

Higher amounts of HSP20 are found in P tissue compared to NP tissue. Similarly, high levels of HSP20 acetylation was found in P tissues and no acHSP20 was present in NP myometrium. This increase in the levels of acetylation might be a result of (i) increase in the levels of protein acetylation or/also (ii) a result of a general increase in HSP20 levels during pregnancy. As acetylation of HSP20 was correlated to myometrial relaxation (2) this results suggest that acetylation of HSP20, exclusively found in pregnant myometrium, may be a contributor to the relaxation of the myometrium prior to labor due to its role in actin remodeling (2).

There is not a lot of knowledge about the KATs which acetylate cytoplasmic proteins. Here we investigated the expression of KAT 9 and CBP to find if they might be putative KATs for HSP20 in human myometrium. Both KAT proteins were detected in NP and P tissue with clearly higher levels in P tissue suggesting their role in increasing protein acetylation levels in P myometrium, however more research need to be done to find if they can acetylate HSP20.

Conclusion

The increase of acHSP20 levels in P myometrium may have a role in keeping the myometrium in a relaxed state during pregnancy.

References

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