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Exploration of the main active metabolites from Tinospora crispa (L.) Hook. f. & Thomson stem as insulin sensitizer in L6.C11 skeletal muscle cell by integrating in vitro, metabolomics, and molecular docking

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Abstract

Ethnopharmacological relevance: Tinospora crispa (L.) Hook. f. & Thomson stem (TCS) has long been used as folk medicine for the treatment of diabetes mellitus. Previous study revealed that TCS possesses multi-ingredients and multi-targets characteristic potential as insulin sensitizer activity. However, its mechanisms of action and molecular targets are still obscure. Aim of the study: In the present study, we investigated the effects of TCS against insulin resistance in muscle cells through integrating in vitro experiment and identifying its active biomarker using metabolomics and in molecular docking validation. Materials and methods: We used centrifugal partition chromatography (CPC) to isolate 33 fractions from methanolic extract of TCS, and then used UHPLC-Orbitrap-HRMS to identify the detectable metabolites in each fraction. We assessed the insulin sensitization activity of each fraction using enzyme-linked immunosorbent assay (ELISA), and then used confocal immunocytochemistry microscopy to measure the translocation of glucose transporter 4 (GLUT4) to the cell membrane. The identified active metabolites were further simulated for its molecular docking interaction using Autodock Tools. Results: The polar fractions of TCS significantly increased insulin sensitivity, as measured by the inhibition of phosphorylated insulin receptor substrate-1 (pIRS1) at serine-312 residue (ser312) also the increasing number of translocated GLUT4 and glycogen content. We identified 58 metabolites of TCS, including glycosides, flavonoids, alkaloids, coumarins, and nucleotides groups. The metabolomics and molecular docking simulations showed the presence of minor metabolites consisting of tinoscorside D, higenamine, and tinoscorside A as the active compounds. Conclusions: Our findings suggest that TCS is a promising new treatment for insulin resistance and the identification of the active metabolites in TCS could lead to the development of new drugs therapies for diabetes that target these pathways. © 2023 Elsevier B.V.

Author keywords

Insulin sensitivity; Menispermaceae; Metabolomics; Natural product; Serine phosphorylation; Type 2 diabetes

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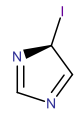

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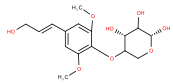

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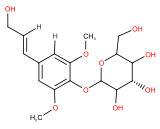

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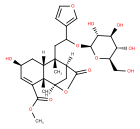

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