SCIENTIFIC REPORT

Project title: Impact of high glucose in valvular endothelial cells-monocyte crosstalk: molecular signatures and role in early valvular dysfunction
Acronym: VALDYSIGN
Phase 3: Evaluation of the function of relevant molecules found to be modified in endothelial permeability and monocyte adhesion and transmigration (part II).
Implementation period: 01.01.2024-31.05.2024
Activity 2.3: Establishing the role of diabetes in monocyte adhesion and transmigration under dynamic conditions.

SUMMARY OF PHASE 3

In this study, our results reveal that the interaction between VEC and monocytes under normal or diabetic conditions has a significant impact on the transcriptomic profile of VEC. Specific molecular changes were identified in processes associated with cytoskeleton regulation, focal adhesion, cell junctions, and cell adhesion molecules. Exposure of VEC to increased glucose concentration led to changes in key molecules and regulatory proteins, such as increased expression of VASP and ROCK1 (involved in cytoskeleton regulation), increased expression of JAM2, cadherin-2, and integrins α4, α5 and β2 (involved in intercellular adhesion and cell-matrix interactions), and decreased expression of paxillin (an adaptive protein in focal adhesions). Additionally, the interaction between VEC and monocytes under normal glucose conditions resulted in increased expression of VASP, ROCK1, integrin β2, and PI3K activation, as well as decreased expression of proteins involved in focal adhesions (FAK and paxillin) and junctional proteins (claudin-5 and cadherin-5). In conditions of increased glucose, this interaction induced PI3K signaling pathway activation, decreased expression of junctional proteins (FAK, caveolin-1, and paxillin), and increased expression of E-selectin. Moreover, the interaction under high glucose conditions highlighted decreased expression of integrins α1, α5, αV, αVβ5, α5β1 and cadherin-2. These changes led to cytoskeleton disorganization, junctional complex alterations, decreased VEC adhesion to extracellular matrix proteins, increased permeability, and increased monocyte adhesion and transmigration, suggesting the alteration of the valvular endothelium's barrier function.
To determine the mechanisms involved in regulating the barrier function, we used Y27623 and LY294002 inhibitors for ROCK and PI3K pathways, identified as activated under diabetic conditions or after monocyte interaction. ROCK and PI3K signaling pathways were identified as activated in VEC under diabetic conditions or after interaction with monocytes. The results show the partial involvement of these signaling pathways in cytoskeleton regulation, regulation of focal adhesion formation or disassembly, intercellular junction regulation, and regulation of cell adhesion molecule expression. Furthermore, PI3K was shown to be involved in regulating endothelial permeability. All of these findings demonstrate that both ROCK and PI3K play an important role in regulating mechanisms induced by increased glucose in the monocyte-VEC interaction and can be potential therapeutic targets in aortic valve disease associated with diabetes.

DISSEMINATION

Publication of a scientific article in an ISI journal - The Specific Molecular Changes Induced

by Diabetic Conditions in Valvular Endothelial Cells and upon Their Interactions with Monocytes Contribute to Endothelial Dysfunction. Monica Madalina Tucureanu, Letitia Ciortan, Razvan Daniel Macarie, Andreea Cristina Mihaila, Ionel Droc, Elena Butoi and Ileana Manduteanu. Int. J. Mol. Sci. 2024, 25,3048. https://doi.org/10.3390/ijms25053048; published on March 6, 2024.