Metabolomic approach reveals potential metabolites for the discrimination of high-grade precancerous lesions of cervical cancer

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Cervical cancer (CC) is one of the malignant tumors that most affect the Brazilian female population. Especially in the state of Amazonas, this neoplasm is the first in incidence and mortality from cancer in women in the state. One of the main ways to prevent this disease is early diagnosis and treatment of pre-cancerous lesions known as cervical intraepithelial neoplasia (CIN), graded from 1 to 3, with stages 2 and 3 being high-grade lesions. These lesions, if not diagnosed and treated early, can progress to CC. Despite different prevention measures have been taken to combat the progression of this disease, incidence and mortality rates in Amazonas remain high. In this context, the use of the metabolomics approach can cooperate in the area of ??oncological studies, where it is possible to identify and analyze possible changes in metabolites that occur in the development of the neoplasia. So, this work aims to characterize the profile of metabolites presents in tissues with high-grade intraepithelial neoplasms. For this, we collected cervix\'s tissues (adjacent margin and the lesion) from patients diagnosed with CIN 2 and CIN 3 (CAAE 71342417.4.3002.502). For metabolomic analysis, tissue samples were subjected to freeze-thaw maceration cycles. The adapted Matyash method was then used for metabolic extraction. Samples were analyzed by LC-HRMS. Multivariate statistics analysis was used to interpret the data obtained, including approaches such as PLS-DA and Random Forest. Finally, MS-DIAL software was used for metabolite identification. In our results, it was observed that there is a significant difference when comparing tissue samples from CIN 2 (lesion and margin) and CIN 3 (lesion and margin). Furthermore, it was possible to evaluate the classification of samples between the CIN 2 and CIN 3 groups using the Random Forest method, which demonstrated predictive accuracy of 88%; thus, allowing the identification of the most important metabolites responsible for the distinction of CIN 2 and CIN 3 samples in this model. One example is methionine sulfoxide, which was overexpressed in CIN 3 samples. This fact is interesting because this metabolite is associated with specific metabolic alterations of carcinogenesis related with the main etiologic agent of these neoplasms, the human papillomavirus (HPV), and therefore may be correlated to a possible evolution of these lesions. In addition, the metabolite L-2-hydroxyglutarate was also identified, which was overexpressed in CIN 2 samples, which has already been associated with carcinogenic processes that may be related to the progression of high-grade CINs. Therefore, we conclude that it was possible to demonstrate the potential of metabolomic analysis in discriminating patients with CIN 2 and CIN 3 using supervised multivariate analyses and the RF predictive model, which may contribute to the emergence of new complementary approaches for the early detection of these neoplasms.

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