Reviewer's report

Title: Mass spectrometry protein expression profiles in colorectal cancer tissue associated with clinico-pathological features of disease

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Reviewer: Lenka Hernychova

Reviewer's report:

Introduced topic is up-to-date. Number of patients suffering with colorectal cancer constantly increase and the development of methods suitable for rapid diagnosis and prognosis estimation are necessary. Colorectal cancer commonly develops through defined stages characterized by molecular alternations and identified biomarkers; therefore each particular stage could be suitable for protein expression profiling. The manuscript describes the recognition of cancer tissue, the discrimination of different stages of the disease, and the prediction of disease recurrence according to the mass spectrometry protein expression profiles. Final data are statistically analyzed and authors try to use the data to create the protein expression profile database.

Major Compulsory Revisions
Regarding high genetic variability of human genome and proteome, the number of analyzed samples (patients in cohort) should be higher.

Minor Essential Revisions
The authors should analyze samples prepared for MALDI-TOF analyses in more detail.
The protein content of representative samples should be identified by advanced mass spectrometric analysis (LC-MS/MS). The differences in protein expression of tumor and normal mucosa tissues should be quantified for example by iTRAQ analyses.

Described method seems to be suitable only for screening and rapid analysis of cancer/normal mucosa tissues. Presented mass spectrometry protein expression profiles are not usable for final validation of result. Nevertheless, this method could be a complementary assay.

The limit of peak detection in linear mass spectrometry mode is not introduced.

Having regard to instrument accuracy, it is arguable whether m/z values could be indicated with one decimal position. The m/z values should be rounded to integers.

The authors should demonstrate representative MALDI mass spectra characteristics for cancer tissue and normal mucosa tissue.
Discretionary Revisions

The authors demonstrate distinction between tumor and normal mucosa tissues based on different protein spectral profiles which are consequently inserted into dataset. For better validation, the team should compare their database system with protein spectral profiles acquired from other samples than those used in the study.

The creation of extensive database containing fingerprint spectra of analyzed tissues is necessary for rapid and screening evaluation of patients’ samples. The database should be able to distinguish positive, negative and poorly characterized samples with statistical significance. This database could be similar to the MALDI Biotyper system, which has been developed for fast and reliable identification of microorganisms.

The classification of clinico-pathological features should be interpreted with caution and final results should be supported with other experimental data (proteomic or genomic).

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:

I declare that I have no competing interests.