Introduction

Pelvic radiotherapy is associated with the occurrence of early and late bladder complications may significantly decrease patient quality of life as frequent, difficult, or painful urination, urinary leakage, nocturia and even blood in urine. Radiobiological mechanisms of complications are known, but little is known about the impact of urinary bacterial flora on the side effects after radiotherapy. Their role in the formation of complications may be essential, as in the case of the gastrointestinal tract, since recent studies suggest that the observed changes are important in the development and prevention of radiation enteropathy. Until recently, any growth of bacteria grown from urine was considered invasive. A new look at the issue of the composition and significance of the urinary microbiome may bring the use of the increasingly popular microorganism mass spectrometry identification method MALDI TOF MS. This technique allows quick and precise identification of microorganisms, although there is still a lack of knowledge on the use of this technique in the microbiome assessment of the human urinary system, especially studies describing the importance of the urinary microbiome in radiation-induced changes.

Aim

The purpose of the study is the development of the MALDI/NALDITOF MS technique for personalized medical diagnosis, with greater emphasis on new sample preparation protocols development for urine bacteria identification as well as investigation of the cases of microorganism's infectious disease its dynamics and evolution in patients with prostate cancer treated with radiotherapy. Particular attention should be paid to the relationship between the composition of bacterial flora and complications after radiation therapy. Therefore, we plan to monitor the bacterial flora in urine collected from patients before the start of radiation therapy and after radiation treatment. Besides, we would like to determine whether there is a relationship between the occurrence of radiation-induced side effects and the pre-treatment bacterial flora and its changes in patients irradiated to the pelvis.

Materials and methods

Prostate cancer patients radically irradiated primary or postoperatively will be included in the study. Treatment planning and treatment will be carried out at the Maria Sklodowska-Curie National Research Institute of Oncology Gliwice Branch, Poland following the current protocol. The clinical side effects of radiation treatment will be assessed using the following questionnaires QLQ 30, QLQ-PR25, EORTC/RTOG. Two urine samples and blood will be collected before the gold fiducial implantation (fiducial is implanted into the prostate gland for precise set up irradiation), on the day of beginning radiotherapy and the last day of radiation treatment, and during check-up 1, 4 months after radiotherapy for analysis. Blood will be used for test analysis, one urine sample will be used for a general urine test and standard urine culture which are routinely carried out during radiotherapy. The second urine sample will be sent to the Interdisciplinary Centre of Modern Technologies Nicolaus Copernicus University in Toruń to perform identification of microorganisms in urine samples of patients using the MALDI/NALDITOF MS. To compare and evaluate the accuracy of performed MALDI identification, isolated bacteria will be also identified and analyzed using reference biochemical tests via automated system BD Phoenix M50^{TM}. The performed classification of the detected bacterial species will be proved using 16S rDNA sequencing. The assessment will include the share of individual groups of microorganisms at the class, government, family, genus and species levels depending on the patient; change in the share of individual groups of microorganisms under the influence of radiation therapy. In addition, a comparison of microorganisms identification results obtained using various techniques for analyzing the microbiological composition of samples and assessments of their usefulness in routine clinical work will be carried out, as well as a selection of optimal conditions for isolation and culture of microorganisms inhabiting the urinary tract. It is planned to perform various techniques of analysis, including the use of multivariate statistical analysis to examine the variability of bacterial composition between patients, as well as changes during radiation therapy.

Expected therapeutic and cognitive benefits

Determination of the microflora from collected urine using molecular and spectrometric tests and correlating it with the occurrence of urinary tract reactions may allow the selection of patients who are more likely to develop urinary complications after radiation therapy. This group of patients should be monitored more closely during radiotherapy to initiate earlier the treatment. In the future, the information obtained may allow assessing the use of prebiotics and probiotics in this group of patients to restore beneficial bacterial flora before radiation treatment, which can help to reduce the side effects of radiotherapy and improve the quality of life. If we are able to select these patients using molecular tests, we intend to correlate the results with routinely performed tests (general urine test and culture) in order to determine the potential parameters that may help to identify the patients at high risk of complications.