

GRAFT QUALITY ASSESSMENT IN KIDNEY TRANSPLANTATION BY MONITORING LIPIDOMIC CHANGES IN THE ORGAN DURING TRANSPLANTATION USING SOLID PHASE MICROEXTRACTION (SPME)

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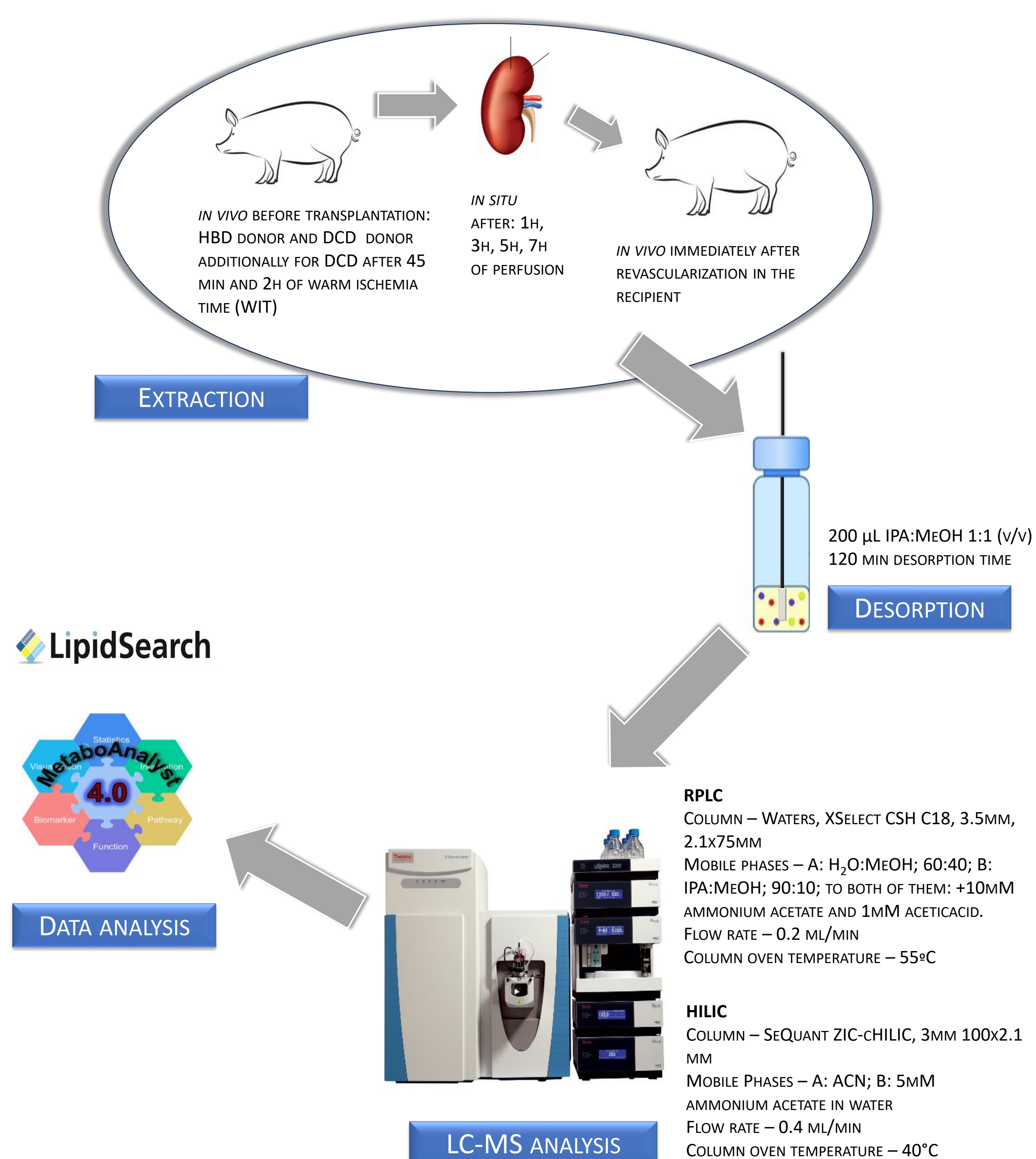
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INTRODUCTION

TRANSPLANTOLOGY IS ONE OF THE FASTEST-GROWING AREA OF MEDICINE AND, UNDOUBTEDLY ORGAN TRANSPLANTATION IS A LIFE-SAVING TREATMENT FOR MILLIONS OF PEOPLE WITH END-STAGE ORGAN DYSFUNCTION. OVER THE PAST FEW DECADES, KIDNEY TRANSPLANTATION HAS SIGNIFICANTLY DEVELOPED AND HAS BECOME A STANDARD PROCEDURE. HOWEVER, IT FACES MANY SERIOUS PROBLEMS, SUCH AS ORGAN SHORTAGE OR LACK OF EFFECTIVE TOOLS FOR ORGAN QUALITY ASSESSMENT. SINCE TISSUE BIOPSY IS INVASIVE AND MACROSCOPIC VISUAL ASSESSMENT IS UNRELIABLE, NEW TECHNOLOGIES ARE STRONGLY NEEDED. SOLID PHASE MICROEXTRACTION (SPME) IS A TECHNOLOGY ALREADY VALIDATED IN MANY DIFFERENT APPLICATIONS IN BIOANALYSIS INCLUDING *EX VIVO* AND *IN VIVO* SAMPLING. THIS METHOD OFFERS SEVERAL ADVANTAGES OF *IN VIVO* TISSUE SAMPLING, SUCH AS LOW INVASIVENESS, EXTRACTION OF UNSTABLE SPECIES, AND LOWER CONSUMPTION OF ORGANIC SOLVENTS. DUE TO THE SMALL SIZE OF THE SPME PROBE, IT IS POSSIBLE TO PERFORM SO CALLED CHEMICAL BIOPSY, WHICH ENABLES EXTRACTION OF SMALL MOLECULES DIRECTLY FROM THE ORGAN WITHOUT ANY TISSUE COLLECTION. **THE AIM OF THIS STUDY WAS TO TEST SPME PROBES AS A LOW INVASIVE TOOL FOR MONITORING LIPIDOMIC CHANGES IN THE ORGAN DURING TRANSPLANTATION.**

METHODS

SPME WAS USED FOR DIRECT KIDNEY SAMPLING AND AS A SAMPLE PREPARATION METHOD. THE STUDY WAS PERFORMED ON KIDNEYS FROM TWO TYPES OF PORCINE MODEL DONORS: HEART BEATING DONOR (HBD) AND DONOR AFTER CARDIAC DEATH (DCD). EXTRACTION WAS EXECUTED VIA SPME PROBE COATED WITH A 7MM MIX-MODE SORBENT.



RESULTS



FIG. 1. PROFILES OF SUMMARY AREA FOR SELECTED CLASSES OF LIPIDS. THE OBTAINED RESULTS INDICATE THAT LEVELS OF LIPIDS FROM THE GLYCEROPHOSPHOLIPID CATEGORY DECREASED DURING PERFUSION. GLYCEROPHOSPHOLIPIDS ARE INVOLVED IN A NUMBER OF PATHOLOGIC PROCESSES BUT MAY ALSO PLAY A PROTECTIVE ROLE AGAINST OXIDATIVE STRESS. DECREASED LEVELS OF THESE LIPIDS MAY REFLECT THEIR CONSUMPTION DURING PERFUSION DUE TO INCREASED ROS PRODUCTION.

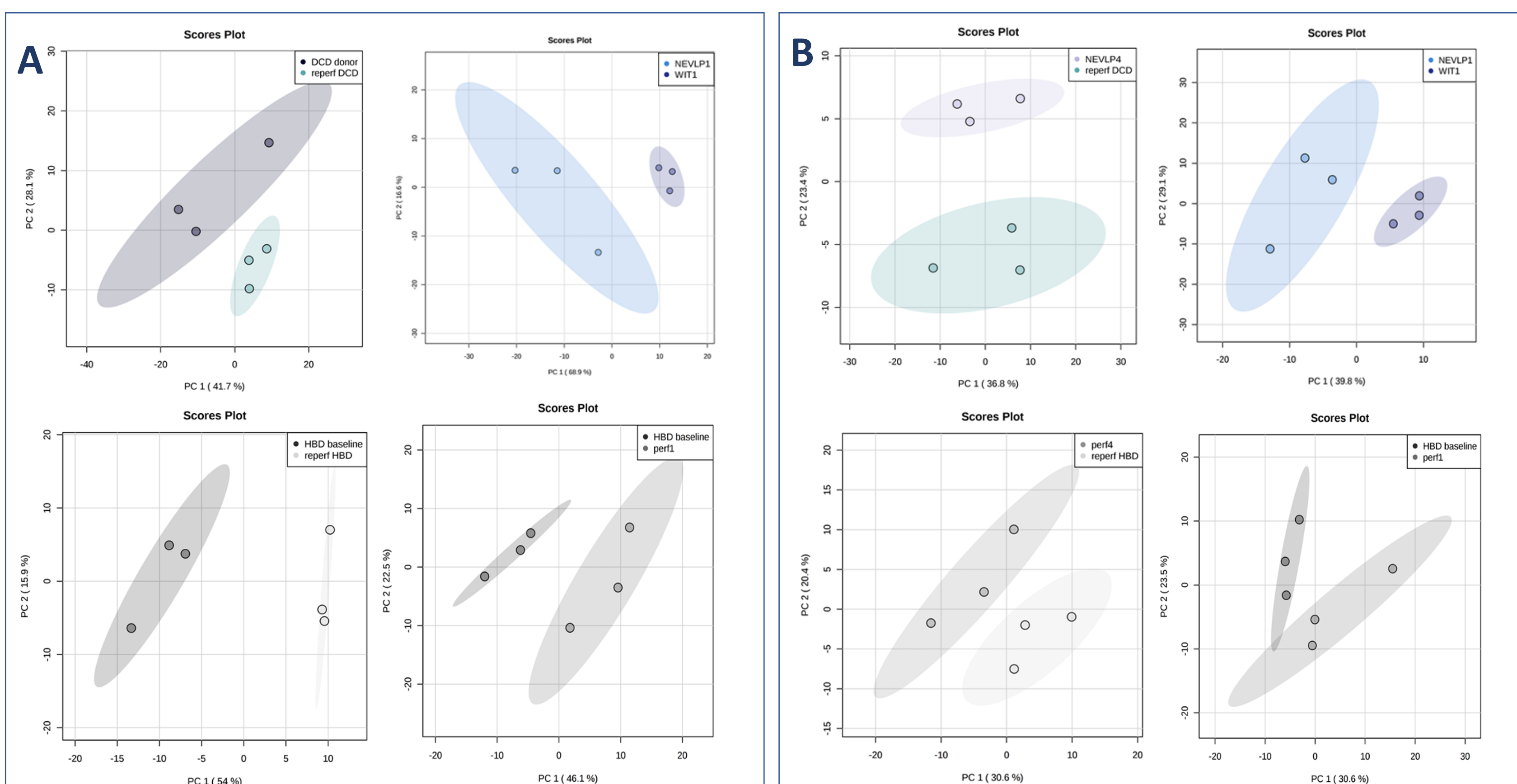


FIG. 2. PRINCIPAL COMPONENT ANALYSIS REPRESENTING CHANGES OCCURRING DURING THE EXPERIMENT. A: HILIC; B: RPLC. THE OBTAINED RESULTS SHOWED DIFFERENCES BETWEEN SAMPLES COLLECTED BEFORE AND AFTER KIDNEY TRANSPLANTATION. IMPORTANTLY, THE OBSERVED DIFFERENTIATION SUGGESTS THAT SPME MAY BE A USEFUL TOOL FOR MONITORING LIPIDOMIC CHANGES THROUGHOUT THE PROCEDURE.

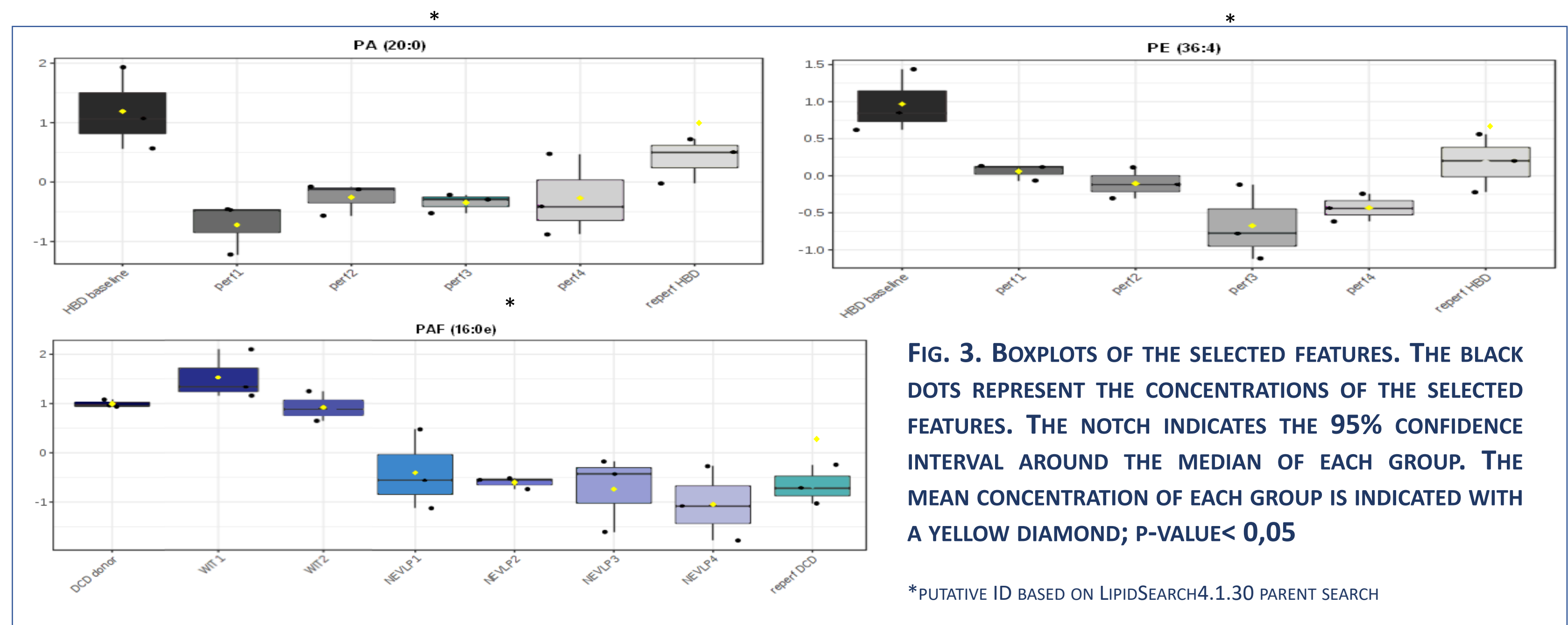


FIG. 3. BOXPLOTS OF THE SELECTED FEATURES. THE BLACK DOTS REPRESENT THE CONCENTRATIONS OF THE SELECTED FEATURES. THE NOTCH INDICATES THE 95% CONFIDENCE INTERVAL AROUND THE MEDIAN OF EACH GROUP. THE MEAN CONCENTRATION OF EACH GROUP IS INDICATED WITH A YELLOW DIAMOND; P-VALUE < 0,05

*PUTATIVE ID BASED ON LIPIDSEARCH4.1.30 PARENT SEARCH

CONCLUSIONS

- THE RESULTS SUGGEST THAT LOW INVASIVE SPME TISSUE SAMPLING, SO CALLED CHEMICAL BIOPSY, MAY PROVIDE IMPORTANT INFORMATION ABOUT BIOCHEMICAL STATUS OF THE GRAFT
- RECEIVED RESULTS INDICATED THAT METABOLITES RELATED TO LIPID METABOLISM MAY BE IMPORTANT IN STUDY OF ISCHEMIA/REPERFUSION GRAFTS INJURY
- THE OBSERVED LIPIDOMIC ALTERATIONS SHOULD BE CONFIRMED IN FURTHER EXPERIMENTS ON LARGER COHORT
- AFTER EXPENSION OF THE STUDY AND VALIDATION OF THE RESULTS THE PRESENTED APPROACH CAN BE FORSEEN AS ADJUNCT DIAGNOSTIC TOOL TO STANDARD PROTOCOL OF GRAFT QUALITY ASSESSMENT IN THE FUTURE

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