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# Methods for studying the bacterial urinary microbiome of non-menopausal women

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## Background

## Objectives

Next generation sequencing technologies (NGS) as well as the development of extended cultures (EC) have allowed the recent discovery of an **Urinary Microbiota (UM)**<sup>1</sup>. Nevertheless, there are still many unknowns regarding the UM associated with healthy subjects or **urinary tract infections (UTIs)**.

- Locally implement **EC** and **NGS** (Illumina and Nanopore) to describe **female bacterial UM**
- Identify **urotype** (dominant bacterial taxon) for each urine specimen
- Compare bacterial UM of patients with **UTIs** with that of **healthy volunteers (HV)**

## Methods

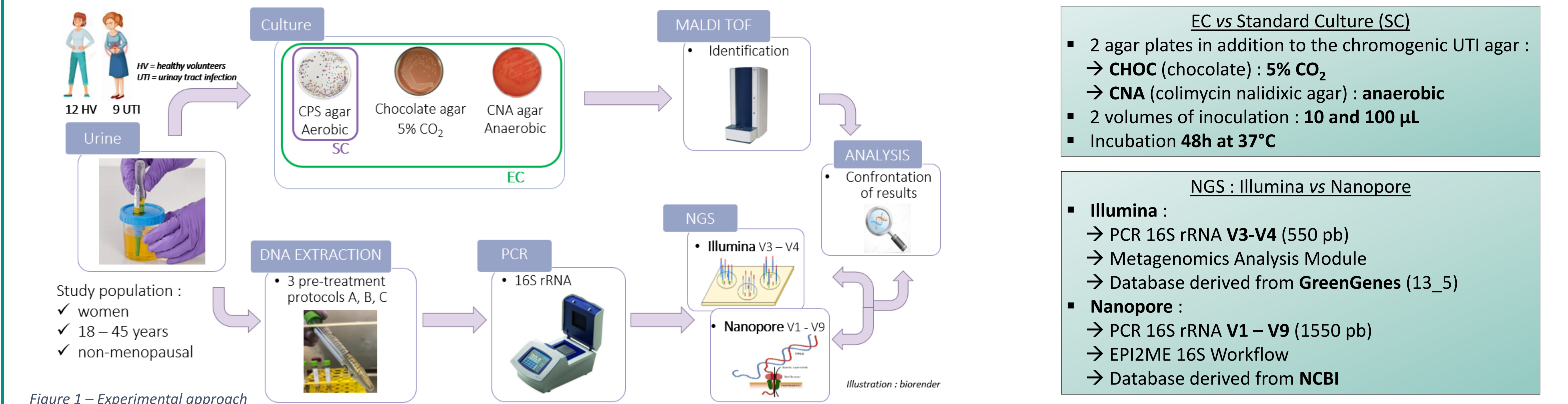


Figure 1 – Experimental approach

## Results

### Analysis of UM by culture

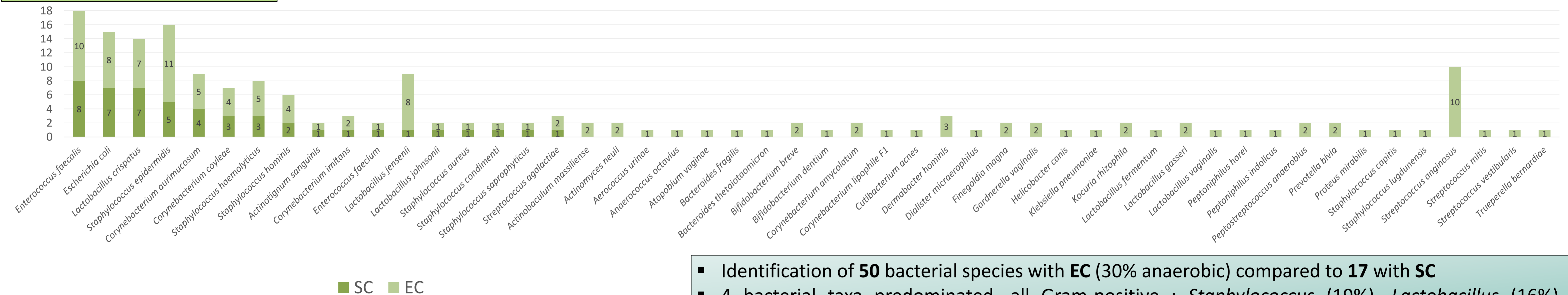


Figure 2 – Prevalence of species identified in standard culture (SC) and in extended culture (EC)

### Analysis of UM by NGS

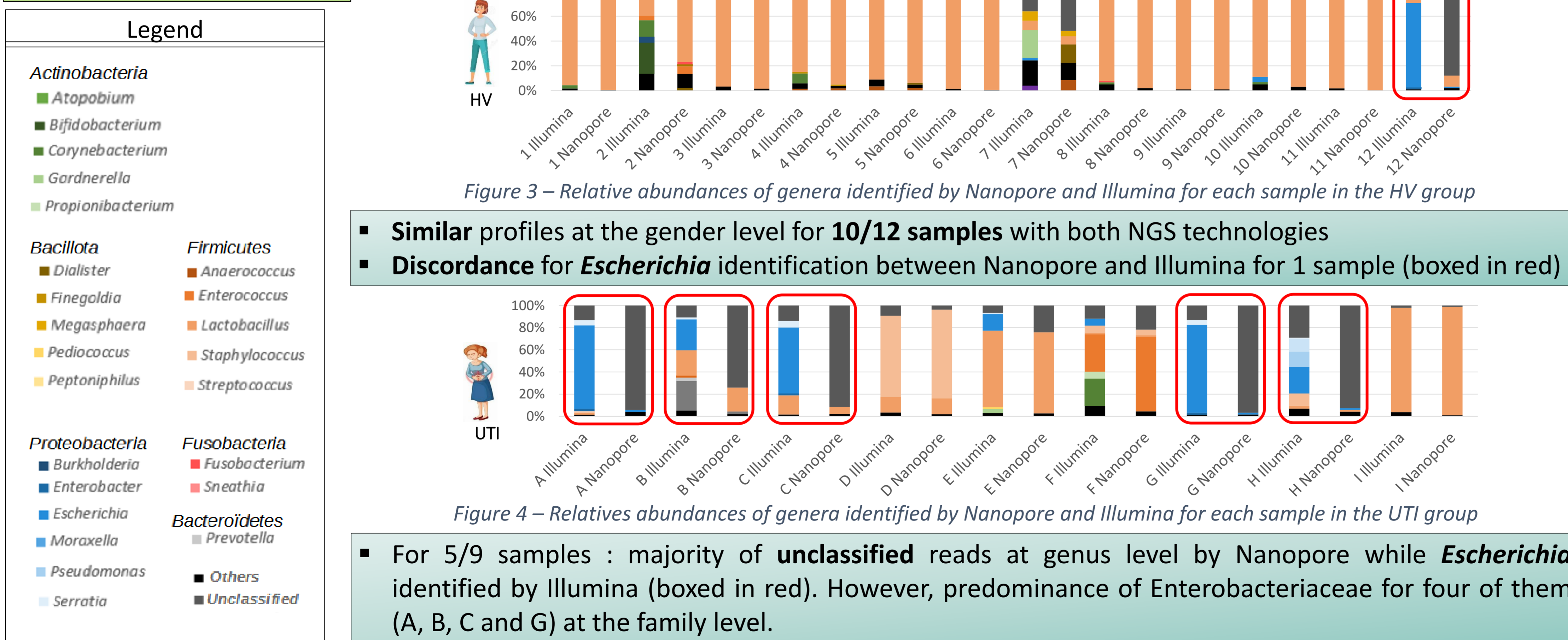


Figure 4 – Relative abundances of genera identified by Nanopore and Illumina for each sample in the UTI group

### UM of UTI vs HV

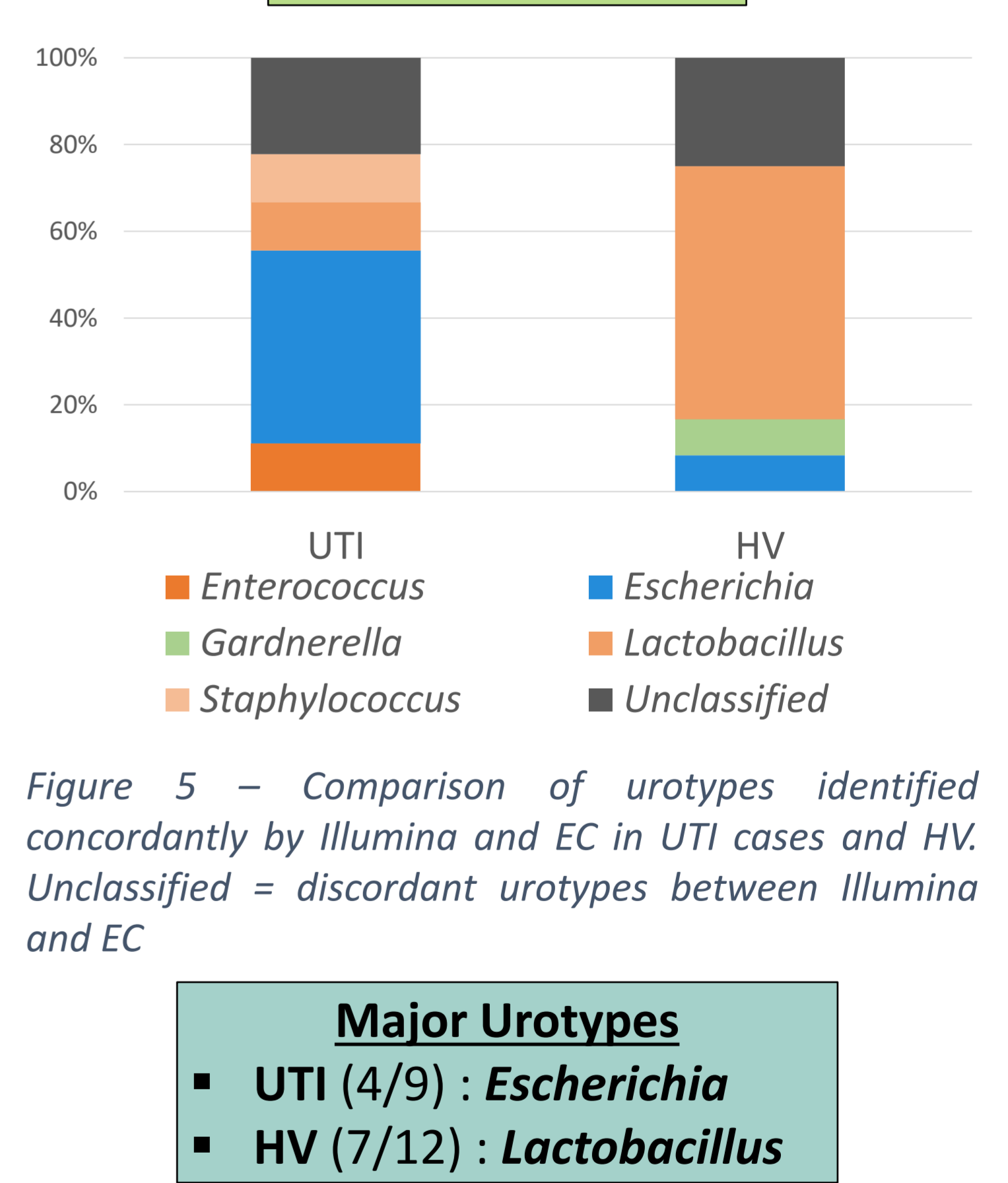


Figure 5 – Comparison of urotypes identified concordantly by Illumina and EC in UTI cases and HV. Unclassified = discordant urotypes between Illumina and EC

## Conclusions

- EC** improves detection of uropathogens compared to SC procedure<sup>1</sup>. Both **EC** and **NGS** results showed a **difference in the composition of the UM** between **HV** and **UTI** patients. This study highlights a high occurrence of ***Lactobacillus*** in the female UM, as already reported<sup>2</sup>.
- Nevertheless, these results must to be confirmed with a larger sample collection, and the analyses of the NGS data need to be further investigated, particularly by using other databases than those suggested by the manufacturers.



1. Price, Travis K., Tanaka Dune, Evann E. Hilt, Krystal J. Thomas-White, Stephanie Kliethermes, Cynthia Brincat, Linda Brubaker, Alan J. Wolfe, Elizabeth R. Mueller, and Paul C. Schreckenberger. "The Clinical Urine Culture: Enhanced Techniques Improve Detection of Clinically Relevant Microorganisms." *Journal of Clinical Microbiology* 54, no. 5 (May 2016): 1216–22.  
 2. Ugarcina Perovic, Svetlana, Magdalena Ksiezarek, Joana Rocha, Elisabete Alves Cappelli, Márcia Sousa, Teresa Gonçalves Ribeiro, Filipa Grosso, and Luísa Peixe. "Urinary Microbiome of Reproductive-Age Asymptomatic European Women." *Theis. Microbiology Spectrum* 10, no. 6 (December 21, 2022): e01308-22.