# Urinary Pathogen ID/ AMR Panel

Highly sensitive, culturefree identification and quantification of common and underrecognized uropathogens

- Identification of > 170 pathogens associated with complicated and uncomplicated infections
- Coverage of > 3700 genes and gene variants associated with antimicrobial resistance
- Compatible with select human and environmental sample types

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# Genetic identification of urinary tract infections

Urinary tract infection (UTI) is any infection occurring in the urinary system (ie, kidneys, ureters, bladder, and urethra). They are among the most common bacterial infections, but UTIs can also be caused by fungal, viral, and parasitic agents. Most infections involve the lower urinary tract, including the bladder and the urethra. Bacterial infections are mostly treated with antibiotics but the rise in antibiotic resistance is a major health concern.<sup>1</sup>

The Urinary Pathogen ID/AMR Panel is a research panel that brings the power of precision metagenomics to uropathogen identification. Panel content is developed based on reported correlation with uncomplicated or complicated UTIs and multidrug-resistant infections. The panel is able to detect and quantify 174 organisms, including common and less common uropathogens and some sexually transmitted pathogens (Figure 1, Table 1). In addition, the panel covers > 3700 genes and gene variants associated with antimicrobial resistance (AMR).

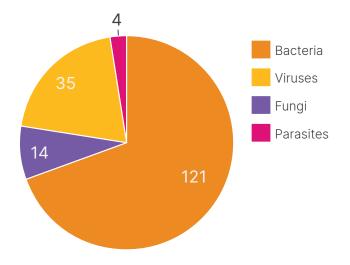


Figure 1: Urinary pathogen categories covered on the Urinary Pathogen ID/AMR Panel.

## Workflow

The Urinary Pathogen ID/AMR Panel enriches for pathogen genomes from a range of host and environmental samples. The workflow includes DNA isolation, library preparation, sequencing, and secondary next-generation sequencing (NGS) analysis (Figure 2). The library preparation and sequencing steps can be completed in two days on benchtop sequencing systems.

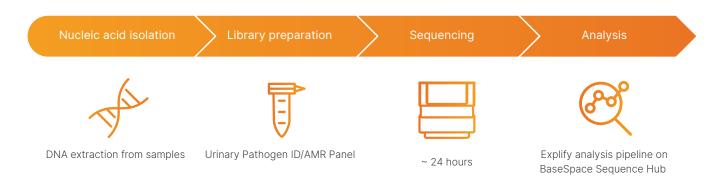


Figure 2: Urinary Pathogen ID/AMR Panel workflow—In a streamlined, comprehensive workflow, libraries are prepared from environmental or host samples, sequenced on any Illumina benchtop sequencing system, and analyzed in the Explify pipeline for pathogen identification, consensus sequence generation, read mapping to pathogen best match, and strain typing. Sequencing time varies with sample read depth and sequencing system used.

#### Table 1: Microorganisms covered on the Urinary Pathogen ID/AMR Panel.

		Reporti	ng name		
Acidovorax wautersii	Candida lusitaniae (Clavispora lusitaniae)	Finegoldia magna (Peptostreptococcus magnus)	Human papillomavirus type 51	<i>Mycobacterium simiae</i> complex	Schistosoma haematobium
Acinetobacter baumannii	Candida parapsilosis	Francisella tularensis	Human papillomavirus type 52	Mycobacterium szulgai	Serratia marcescens
Acinetobacter calcoaceticus	Candida tropicalis	Fusobacterium necrophorum	Human papillomavirus type 54	Mycobacterium tuberculosis complex	Simian virus 40
Acinetobacter pittii	Chlamydia trachomatis	Fusobacterium nucleatum	Human papillomavirus type 55/44	Mycobacterium xenopi	Sneathia amnii (Leptotrichia amnionii)
Actinobaculum massiliense	Chromobacterium violaceum	Giardia intestinalis	Human papillomavirus type 56	Mycobacteroides chelonae (Mycobacterium chelonae)	Staphylococcus aureus
Actinotignum sanguinis	Chryseobacterium indologenes	Haemophilus ducreyi	Human papillomavirus type 58	Mycoplasma genitalium	Staphylococcus epidermidis
Actinotignum schaalii (Actinobaculum schaalii)	Citrobacter freundii complex	Haemophilus influenzae	Human papillomavirus type 6	Mycoplasma hominis	Staphylococcus haemolyticus
Aerococcus christensenii	Citrobacter koseri	Haemophilus parainfluenzae	Human papillomavirus type 66	Neisseria gonorrhoeae	Staphylococcus hominis
Aerococcus sanguinicola	Corynebacterium aurimucosum	Hafnia alvei	Human papillomavirus type 68	Neisseria meningitidis	Staphylococcus intermedius
Aerococcus urinae	Corynebacterium coyleae	Herpes simplex virus 1	Human papillomavirus type 70	Oligella urethralis	Staphylococcus lugdunensis
Aerococcus viridans	Corynebacterium genitalium	Herpes simplex virus 2	JC polyomavirus	Pantoea agglomerans	Staphylococcus saprophyticus
Aeromonas hydrophila	Corynebacterium glucuronolyticum	Histoplasma capsulatum	Klebsiella aerogenes (Enterobacter aerogenes)	Pasteurella multocida	Staphylococcus simular
Alloscardovia omnicolens	Corynebacterium jeikeium	Human adenovirus B	Klebsiella oxytoca	Peptostreptococcus anaerobius	Staphylococcus warner
Anaerococcus lactolyticus	Corynebacterium pseudogenitalium	Human adenovirus C	Klebsiella pneumoniae	Porphyromonas asaccharolytica	Stenotrophomonas maltophilia
Anaerococcus vaginalis (Peptostreptococcus vaginalis)	Corynebacterium renale	Human adenovirus E	Klebsiella quasipneumoniae	Prevotella timonensis	Streptococcus agalactia
Aspergillus flavus	Corynebacterium riegelii	Human herpesvirus 6	Klebsiella variicola	Propionimicrobium Iymphophilum	Streptococcus anginosu
Atopobium vaginae	Corynebacterium urealyticum	Human herpesvirus 7	Kluyvera ascorbata	Proteus mirabilis	Streptococcus constellatus
Bacillus subtilis	Cryptococcus neoformans	Human herpesvirus 8	Kocuria rosea	Proteus penneri	Streptococcus intermedius
Bacteroides fragilis	Cytomegalovirus	Human papillomavirus type 11	Listeria monocytogenes	Proteus vulgaris	Streptococcus pneumoniae
Bifidobacterium breve	Dialister micraerophilus	Human papillomavirus type 16	Mobiluncus curtisii	Providencia rettgeri	Streptococcus pyogene
3K polyomavirus	Entamoeba histolytica	Human papillomavirus type 18	Mobiluncus mulieris	Providencia stuartii	Treponema pallidum
Burkholderia cepacia complex	Enterobacter cloacae complex	Human papillomavirus type 30	Molluscum contagiosum virus	Pseudomonas aeruginosa	Trichomonas vaginalis
Campylobacter jejuni	Enterococcus faecalis	Human papillomavirus type 31	Moraxella catarrhalis	Pseudomonas mendocina	Trichosporon asahii
Candida albicans	Enterococcus faecium	Human papillomavirus type 33	Morganella morganii	Pseudomonas putida	Trichosporon beigelii (Cutaneotrichosporon cutaneum)
Candida auris (Clavispora auris)	Enterococcus gallinarum	Human papillomavirus type 35	<i>Mycobacterium avium</i> complex	Rahnella aquatilis	Ureaplasma parvum
Candida dubliniensis	Enterococcus raffinosus	Human papillomavirus type 39	Mycobacterium gordonae	Rhodanobacter denitrificans	Ureaplasma urealyticum
Candida fabianii Cyberlindnera fabianii)	Epstein-Barr virus	Human papillomavirus type 42	Mycobacterium kansasii	Riemerella anatipestifer	Veillonella parvula
Candida glabrata (Nakaseomyces glabrata)	Escherichia coli	Human papillomavirus type 43	Mycobacterium marinum	Rothia kristinae (Kocuria kristinae)	Vibrio cholerae
Candida krusei (Pichia kudriavzevii)	Facklamia hominis	Human papillomavirus type 45	Mycobacterium scrofulaceum	Salmonella enterica	Yersinia enterocolitica

#### Library preparation

The Urinary Pathogen ID/AMR Enrichment Kit contains all library prep reagents, indexes, and panel probes for on-bead tagmentation followed by a single hybridization step to provide a rapid workflow for generating enriched libraries. The probe-based enrichment provides:

- Improved sensitivity over shotgun metagenomic sequencing
- Rapid, automation-compatible workflow with minimal hands-on time
- Scalable throughput that supports multiplexing of up to 384 samples in a single run

Learn more about on-bead tagmentation, illumina. com/techniques/sequencing/ngs-library-prep/ tagmentation.html

#### Sequencing

The sensitivity of probe-based target enrichment used with the Urinary Pathogen ID/AMR Panel libraries allows for flexible sequencing options. The panel is ideal for use with the benchtop MiniSeq<sup>™</sup>, MiSeq<sup>™</sup>, and NextSeq<sup>™</sup> 550, NextSeq 1000, and NextSeq 2000 systems. Nucleic acid sample quality, sample read depth, and the number of reads per sample impact the number of pathogen-specific reads and sequence coverage obtained.

#### Data analysis

In order to support various experimental workflows, the Urinary Pathogen ID/AMR Panel includes probes targeting 10 commercially available spike-in control options (Table 2). Analysis is performed with the Explify<sup>™</sup> secondary analysis pipeline, available on BaseSpace<sup>™</sup> Sequence Hub.

The data analysis pipeline provides contig assembly, consensus sequences, and genome coverage metrics for the pathogens featured in the panel. Users can select individual samples or a BaseSpace Sequence Hub project folder containing samples for analysis. The application interface allows for configuration of analysis options, including read quality control metrics, quantitative spike-in control selection, and filtered reporting for AMR markers and microorganisms. Sample results can be output in six different formats: summary PDF report, spreadsheet of results in XLSX format, comprehensive JSON report, interactive HTML web report, annotated TSV variant table with known bacterial AMR marker variants, and consensus genome FASTA sequences for some organisms. Additionally, users can download an aggregate XLSX report for all samples in a BaseSpace Sequence Hub analysis folder to facilitate downstream analysis of results from related samples (eg, results from the same experiment).

#### Table 2: Spike-in control options.

Allobacillus halotolerans

Armored RNA Quant Internal Process Control

Enterobacteria phage T7

Escherichia virus MS2

Escherichia virus Qbeta

Escherichia virus T4

Imtechella halotolerans

Phocid alphaherpesvirus 1

Phocine morbillivirus

Truepera radiovictrix

## Performance

#### Sensitive pathogen detection

The target enrichment design of the Urinary Pathogen ID/AMR Panel is highly sensitive, outperforming shotgun metagenomics and conventional urine culture methods for identifying common uropathogens. The target enrichment approach also allows for greater analytical sensitivity at a lower number of total reads compared to shotgun metagenomics methods, ~1M vs. ~10M, 1 × 147 bp paired-end reads (Figure 3).

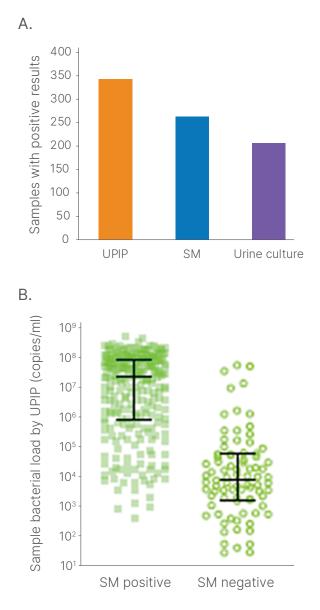


Figure 3: Sensitivity of Urinary Pathogen ID/AMR Panel—The Urinary Pathogen ID/AMR Panel (UPIP) offers more sensitive pathogen identification than shotgun metagenomics (SM) and culture methods. Sequencing was performed on a NextSeq 550 system. (A) One or more uropathogens were detected in 342/399 (86%) urine samples by UPIP, in 262/399 samples (66%) by SM, and in 205/399 samples (51%) by urine culture. (B) Bacterial load by UPIP was higher in SM-positive than in SM-negative samples (p < 0.0001), consistent with the prediction of improved analytical sensitivity of targeted enrichment sequencing compared with SM.

#### Summary

The Urinary Pathogen ID/AMR Panel offers sensitive detection of > 170 common and underrecognized uropathogens. The panel is also able to identify > 3700 antimicrobial resistance markers for the included pathogens, enabling investigations into resistance profiles. The target–capture approach of the Urinary Pathogen ID/AMR Panel means greater detection sensitivity than shotgun metagenomics sequencing methods and makes it ideal for benchtop sequencing systems. The panel is compatible with a range of environmental and biological research sample types.

### Learn more

Urinary Pathogen ID/AMR Panel, illumina.com/products/ by-type/sequencing-kits/library-prep-kits/urinarypathogen-id-amr-enrichment-kit.html

BaseSpace applications, illumina.com/products/bytype/informatics-products/basespace-sequence-hub. html

Illumina sequencing platforms, illumina.com/systems/ sequencing-platforms.html

## Ordering information

Product	Catalog no.
Urinary Pathogen ID/AMR Enrichment Kit Set A (RUO) (96 indexes,96 samples)	20090308
Urinary Pathogen ID/AMR Enrichment Kit Set B (RUO) (96 indexes,96 samples)	20090309
Urinary Pathogen ID/AMR Enrichment Kit Set C (RUO) (96 indexes, 96 samples)	20090310
Urinary Pathogen ID/AMR Enrichment Kit Set D (RUO) (96 indexes, 96 samples)	20090311

## References

 Bono MJ, Leslie SW, Reygaert WC. Urinary Tract Infection. In: StatPearls. Treasure Island (FL): StatPearls Publishing; June 15, 2022.

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