

# cfDNA isolation from first-void urine using the Colli-Pee<sup>®</sup> device containing UAS

Jordaens S.<sup>1,2</sup>, Zwaenepoel K.<sup>1,3</sup>, Vorsters A.<sup>4</sup>, Deben C.<sup>1</sup>, Tjalma W.<sup>5</sup>, De Wachter S.<sup>6</sup>, Van Dam P.<sup>5</sup>, Vankerckhoven V.<sup>2</sup>, Pauwels P.<sup>1,3</sup>

1| Center for Oncological Research (CORE), Integrated Personalized and Precision Oncology Network (IPPON), University of Antwerp, Wilrijk, Belgium 2| Novosanis NV, Research and Development, Wijnegem, Belgium 3| Laboratory of Pathological Anatomy, Antwerp University Hospital (UZA), Edegem, Belgium 4| Center for Evaluation of Vaccination (CEV), Vaccine & Infectious Disease Institute (VAXINFECTIO), University of Antwerp, Wilrijk, Belgium 5| Multidisciplinary Breast Unit, Antwerp University Hospital (UZA), Edegem, Belgium 6| Urology, Antwerp University Hospital (UZA), Edegem, Belgium

## AIM

The aim of this study was to evaluate Colli-Pee<sup>®</sup> containing UAS for cell-free (cf) DNA isolation from first-void urine.

## METHODS

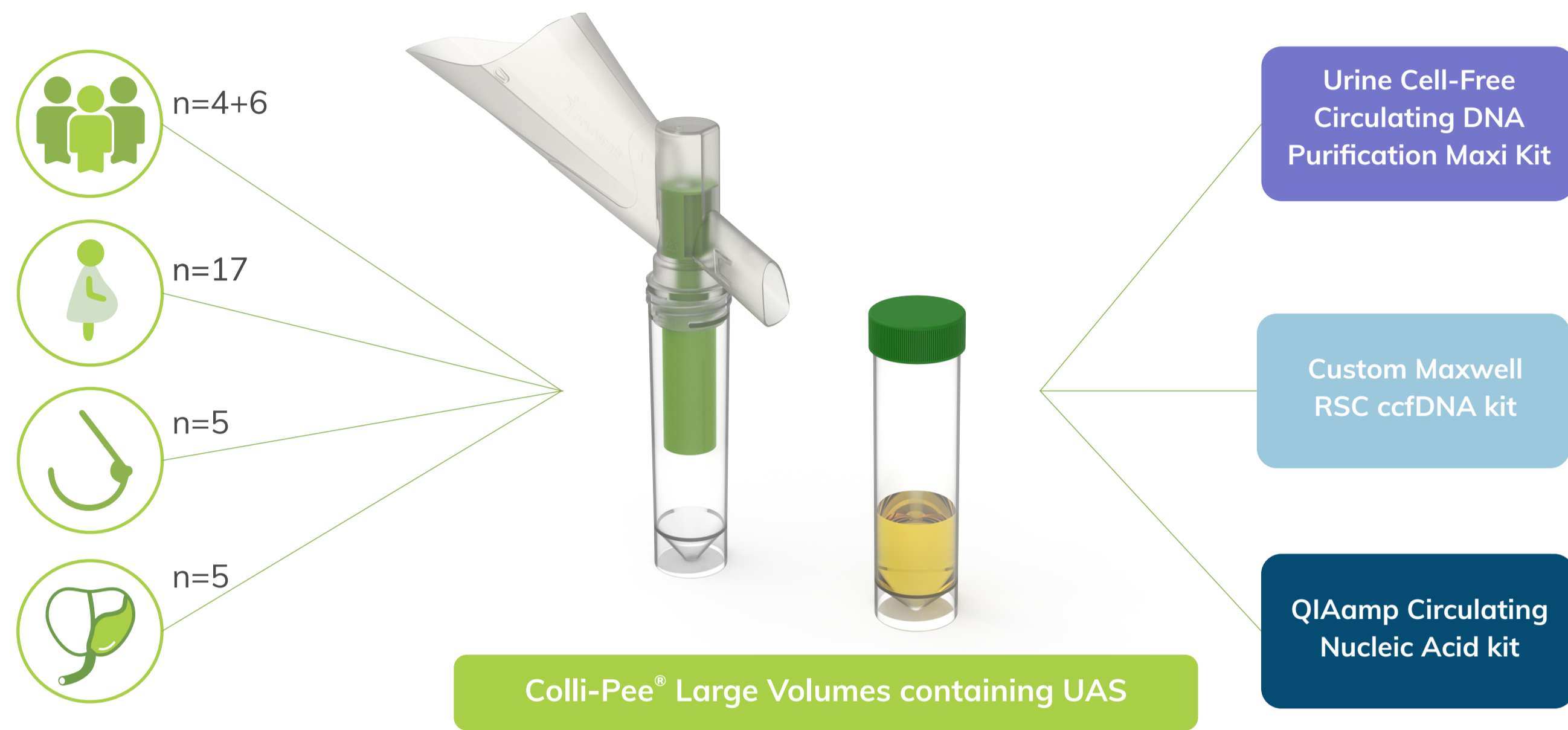


Figure 1: Study set-up

A total of 37 participants (4 female and 6 male healthy volunteers, 17 pregnant women, 5 breast and 5 prostate cancer patients) collected 45mL of urine using the Colli-Pee<sup>®</sup> Large Volumes containing UAS (Novosanis). For the isolation of cfDNA from the UAS-preserved urine samples, 3 commercially available kits were used: QIAamp Circulating Nucleic Acid kit (Qiagen), Urine Cell-Free Circulating DNA Purification Maxi kit (Norgen) and Custom Maxwell RSC cfDNA kit (Promega). The DNA concentration was measured using the Qubit dsDNA HS assay kit and the fragment lengths and percentage of cfDNA were measured using the cfDNA ScreenTape for the TapeStation (Agilent). Usability of the Colli-Pee<sup>®</sup> Large Volumes containing UAS was evaluated through a questionnaire.

## RESULTS

### DNA QUANTITY AND QUALITY:

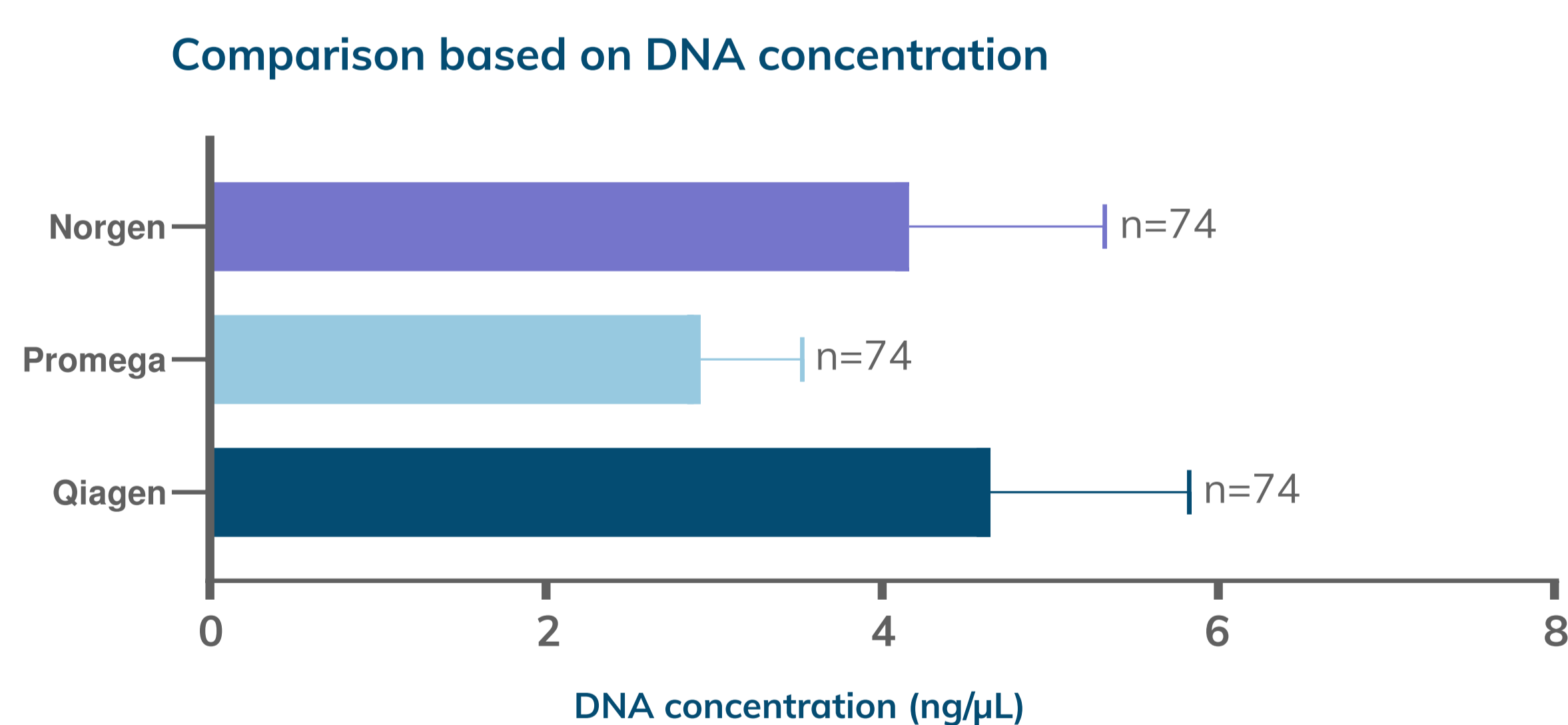


Fig 2. Comparison isolation methods based on DNA concentration. Depicted as mean ± SEM

Qubit results (Fig 2.) showed a larger DNA concentration when isolation was performed using the Norgen (c=4.161 ng DNA/μL) or the Qiagen (c=4.645 ng DNA/μL) kit, while the Promega kit provided a lower concentration (c=2.922 ng DNA/μL).

### Comparison isolation methods based on DNA concentration per participant type

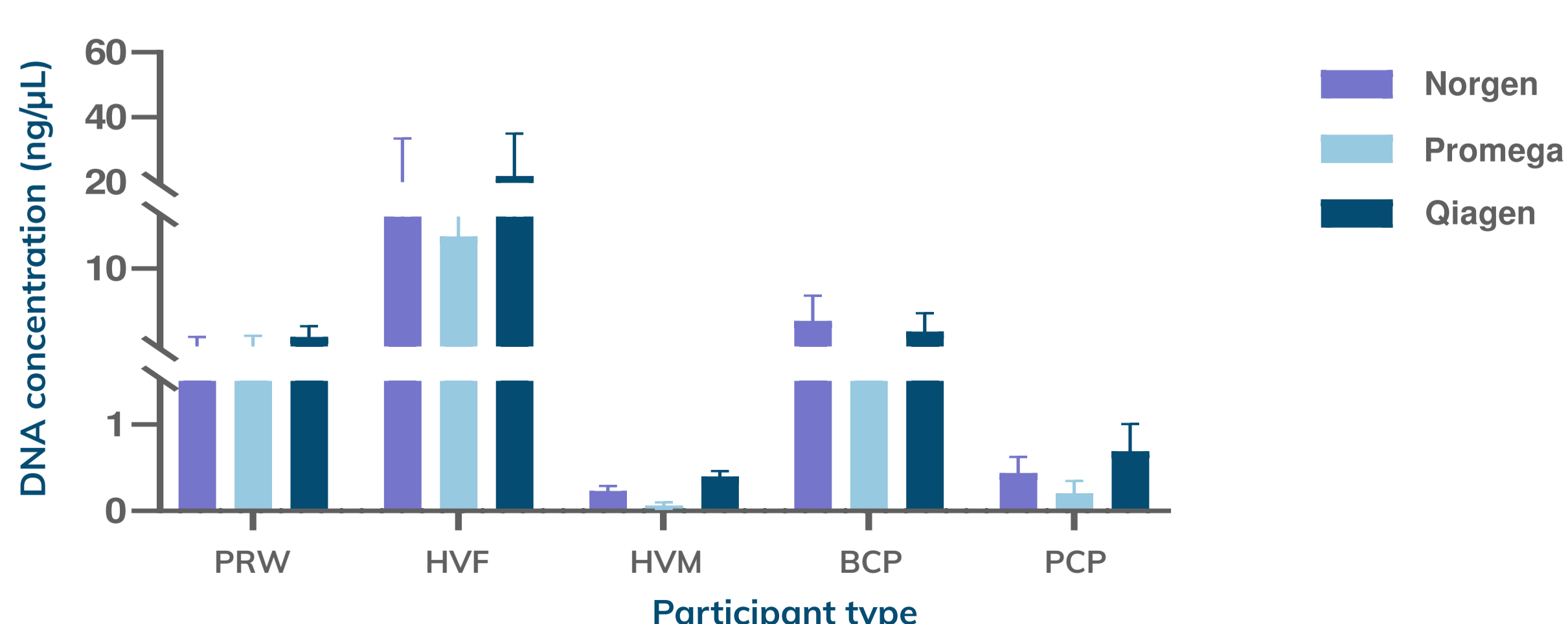


Figure 3: Comparison isolation methods based on DNA concentration per participant type. Depicted as mean ± SEM, BCP: breast cancer patients; HVF: healthy female volunteers; HVM: healthy male volunteers; PCP: prostate cancer patients; PRW: pregnant women

It is clearly seen (Fig 3.) that the concentration of DNA is lower in urine from male participants (i.e., healthy male volunteers and prostate cancer patients) than from female participants (i.e., pregnant women, healthy female volunteers and breast cancer patients).

### Comparison based on cfDNA percentage per participant type

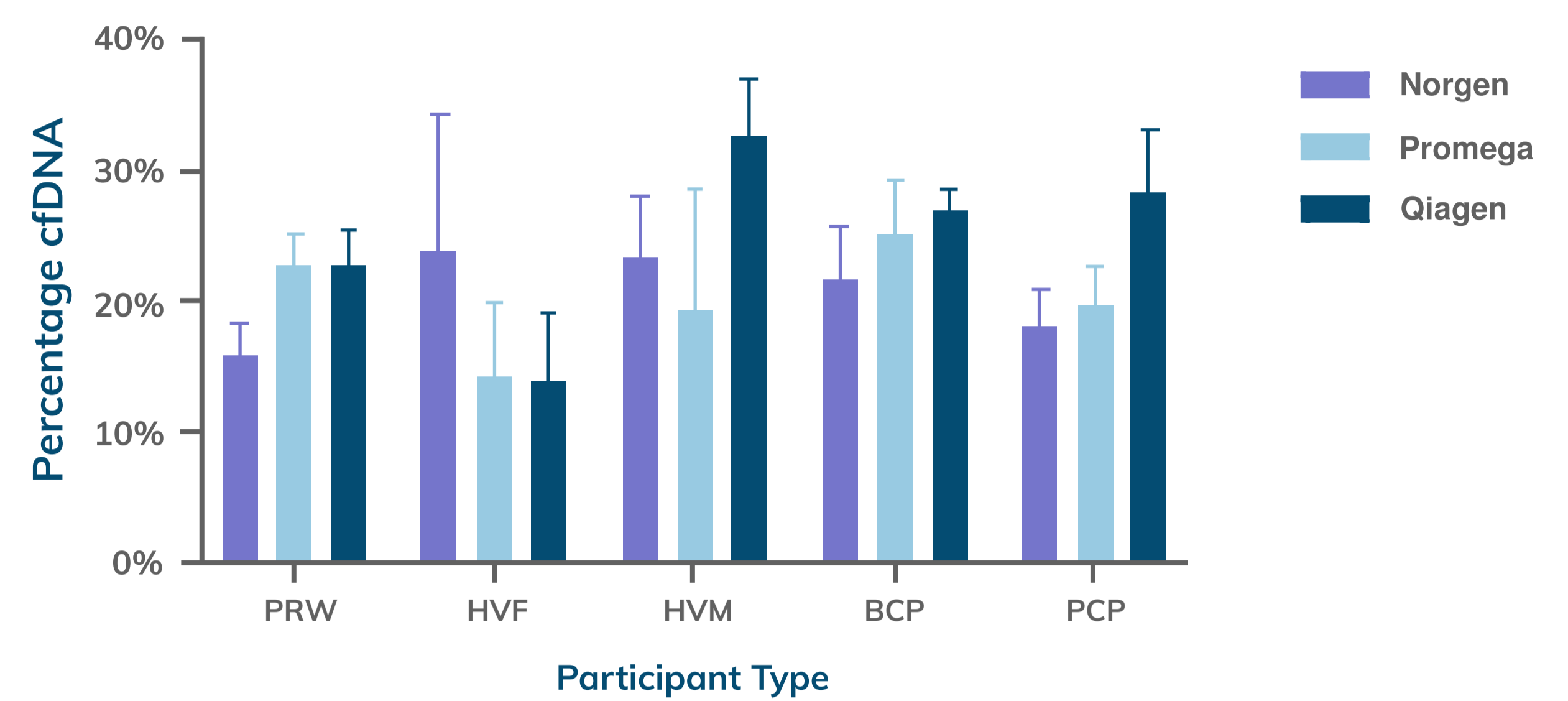
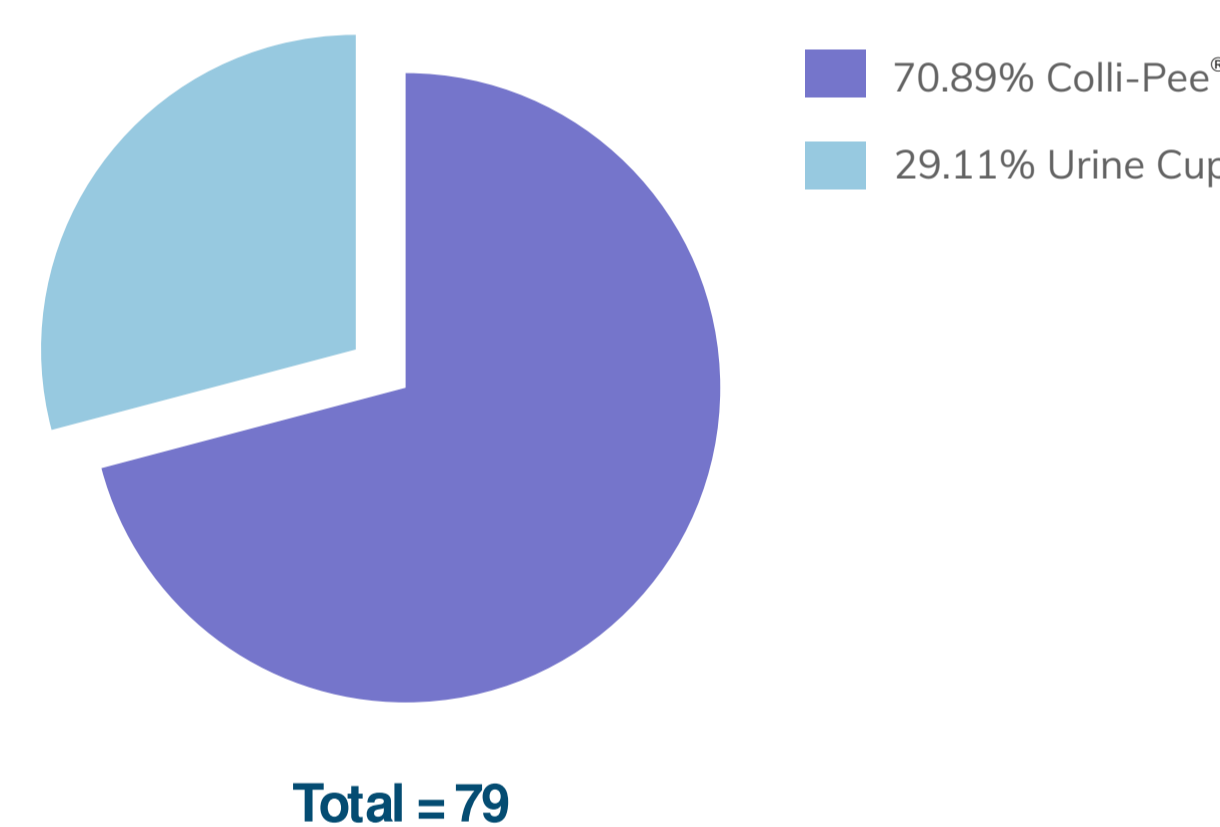


Fig 4. Comparison isolation methods based on cfDNA percentage per participant type. Depicted as mean ± SEM, BCP: breast cancer patients; HVF: healthy female volunteers; HVM: healthy male volunteers; PCP: prostate cancer patients; PRW: pregnant women

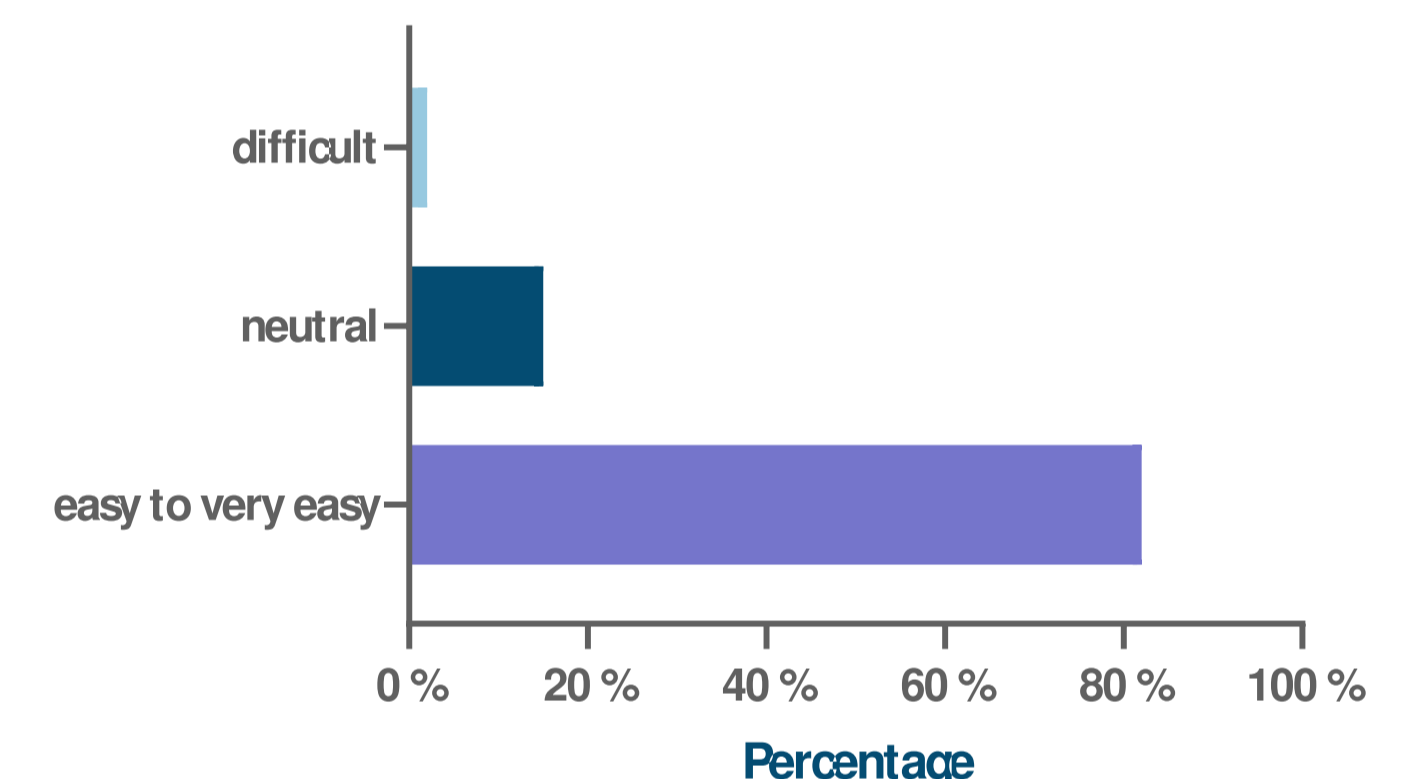
Using TapeStation, percentages of cfDNA (50-450 bp) were determined to be around 20% independent of the isolation method or participant type.

### USABILITY:

#### Preferred collection method



#### General Experience Colli-Pee<sup>®</sup>



Questions	Score on 100*				
	< 50	50-79	80-99	100	
Before collection	It was clear how to build Colli-Pee <sup>®</sup>	1%	8%	35%	56%
	Mounting tube on Colli-Pee <sup>®</sup> was easy	1%	6%	37%	63%
	The montage of Colli-Pee <sup>®</sup> was fast	1%	2%	34%	63%
During collection	IFU was sufficient to use Colli-Pee <sup>®</sup>	1%	12%	25%	62%
	It was clear how to use Colli-Pee <sup>®</sup>	0%	10%	37%	53%
After collection	It was comfortable to use Colli-Pee <sup>®</sup>	5%	27%	35%	32%
	Demounting the tube was easy	0%	9%	37%	54%
	Closing the tube with cap was easy	0%	3%	30%	67%

\*Number of participants that completed the questions ranged from 89 to 97 participants.

Figure 5: Usability results summary

Until August 2021, almost 100 participants completed the URODETECT questionnaire. The ages of the participants ranged from 19 to 83 years. Almost all participants rated the usability of Colli-Pee<sup>®</sup> Large Volumes, before, during and after collection higher than 80 (on 100). In addition, they also rated the general usability of Colli-Pee<sup>®</sup> Large Volumes as easy to very easy. And 71% preferred Colli-Pee<sup>®</sup> over a regular urine cup (29%) for the collection of urine.

## CONCLUSION

All three commercially available cfDNA isolation kits were able to isolate the cfDNA from the UAS-preserved urine samples. Results were comparable, although Qiagen seemed the better fit. Colli-Pee<sup>®</sup> Large Volumes shows good usability results, which will be considered while further improving the Colli-Pee<sup>®</sup> Large Volumes.