Comparison of three commercially available kits for cfDNA isolation from First-Void urine collected with Colli-Pee® containing UAS

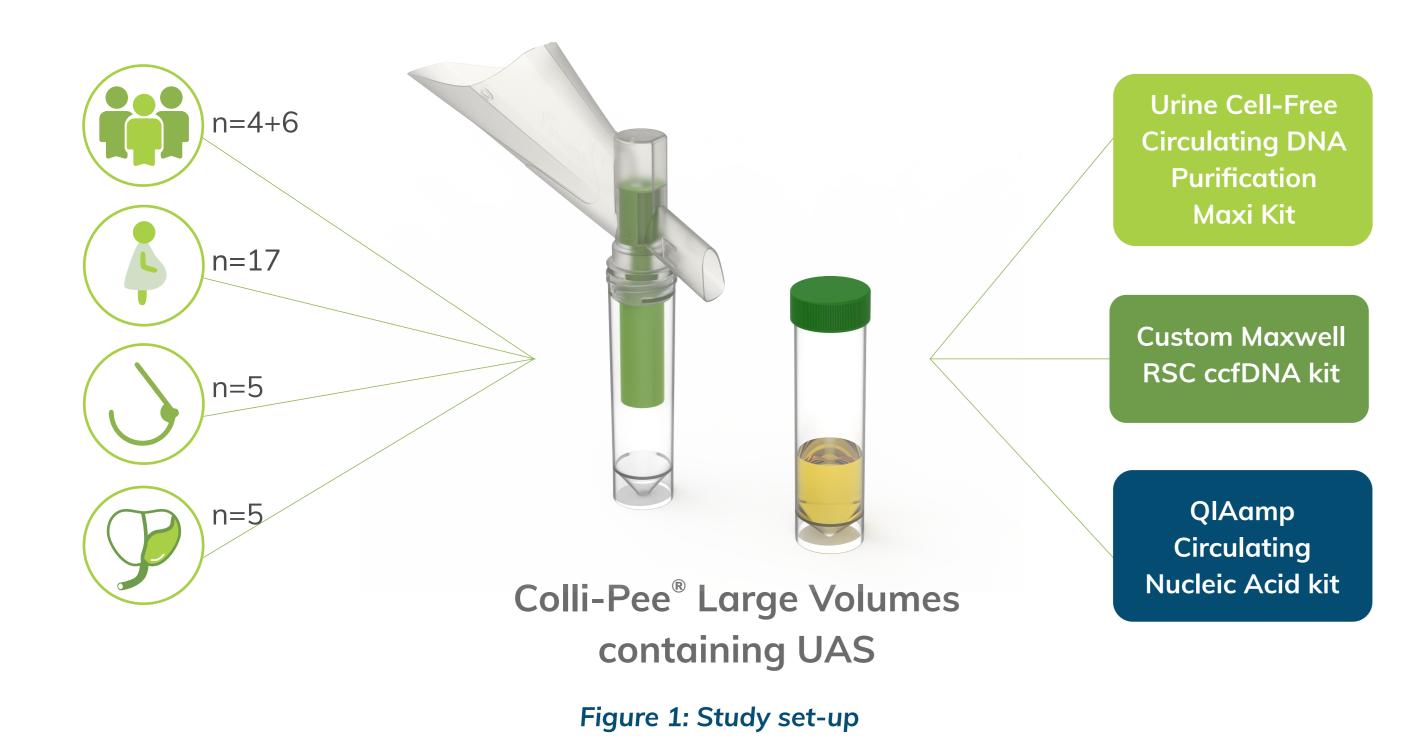
Jordaens S.^{1,2}, Zwaenepoel K.^{1,3}, Vorsters A.⁴, Beyers, K.², Deben, C.¹, Tjalma, W.⁵, De Wachter S.⁶, Van Dam P.⁵, Vankerckhoven V.², Pauwels P^{1,3}.

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

AIM

The aim of this study was to compare different commercially available kits for cell-free (cf) DNA isolation from first-void urine (FVU).

METHODS



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[®] Large Volumes containing UAS* (Novosanis). cfDNA was isolated from FVU using 3 commercially available kits: QIAamp Circulating Nucleic Acid kit (Qiagen), Urine Cell-Free Circulating DNA Purification Maxi kit (Norgen) and Custom Maxwell RSC ccfDNA kit (Promega).

The DNA concentration was measured using the Qubit dsDNA HS assay kit and the fragment lengths and percentage of cfDNA was measured using the cfDNA ScreenTape for the TapeStation (Agilent). Usability of the Colli-Pee® Large Volumes containing UAS* was evaluated through a questionnaire.

RESULTS

The three cfDNA isolation kits showed small method variance, while a larger variance was seen between samples. Qubit results showed a larger DNA concentration when isolation was performed using the Qiagen (c=4.645 ng DNA/µL) or Norgen (c=4.161 ng DNA/µL) kit, while Promega provided a lower concentration (c=2.922 ng DNA/µL). A non-significant trend towards lower cfDNA concentration was noted in urine from male participants than from female participants.

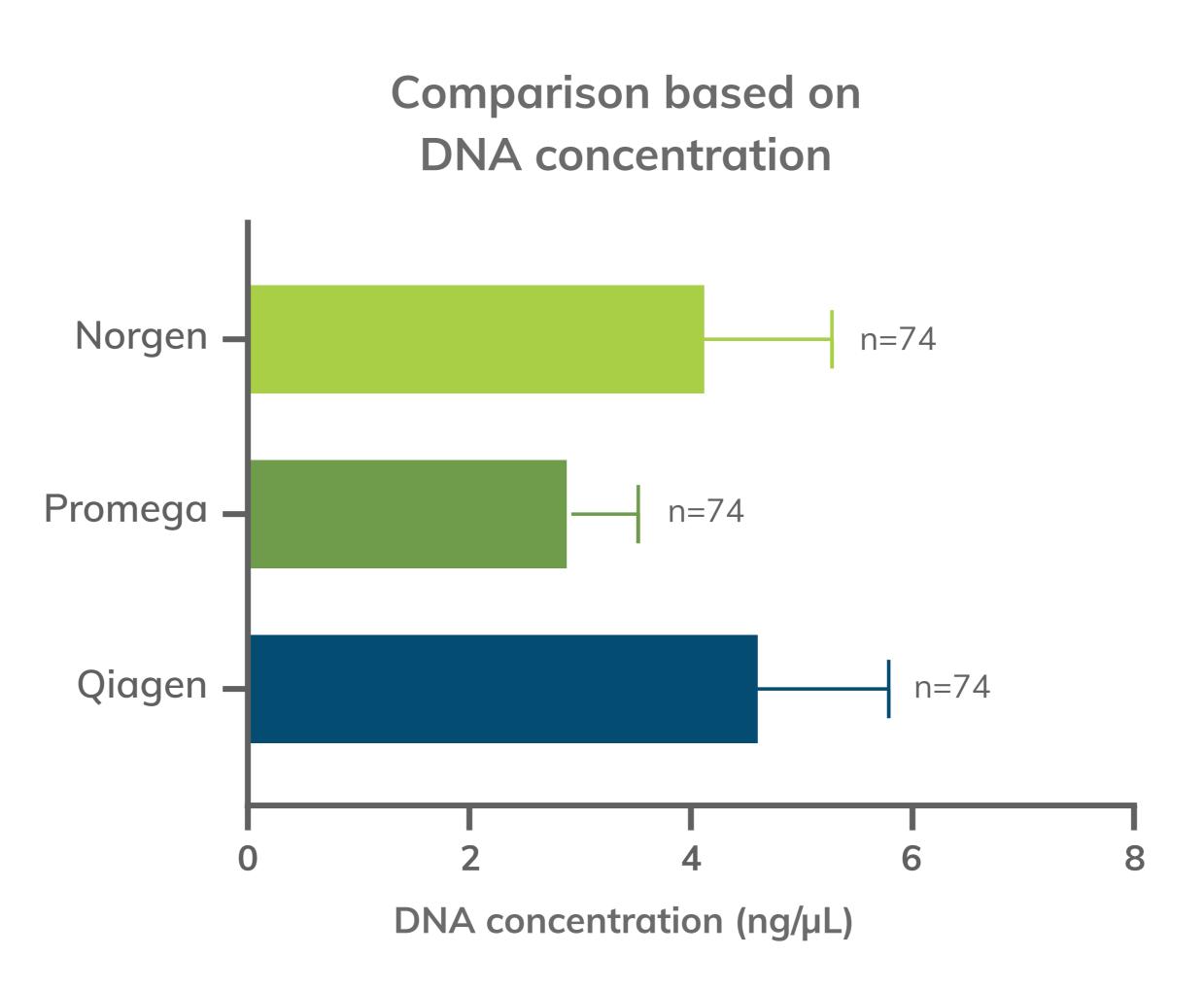


Fig 2. Comparison isolation methods based on DNA concentration Depicted as mean \pm SEM

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

Comparison based on cfDNA percentage per participant type

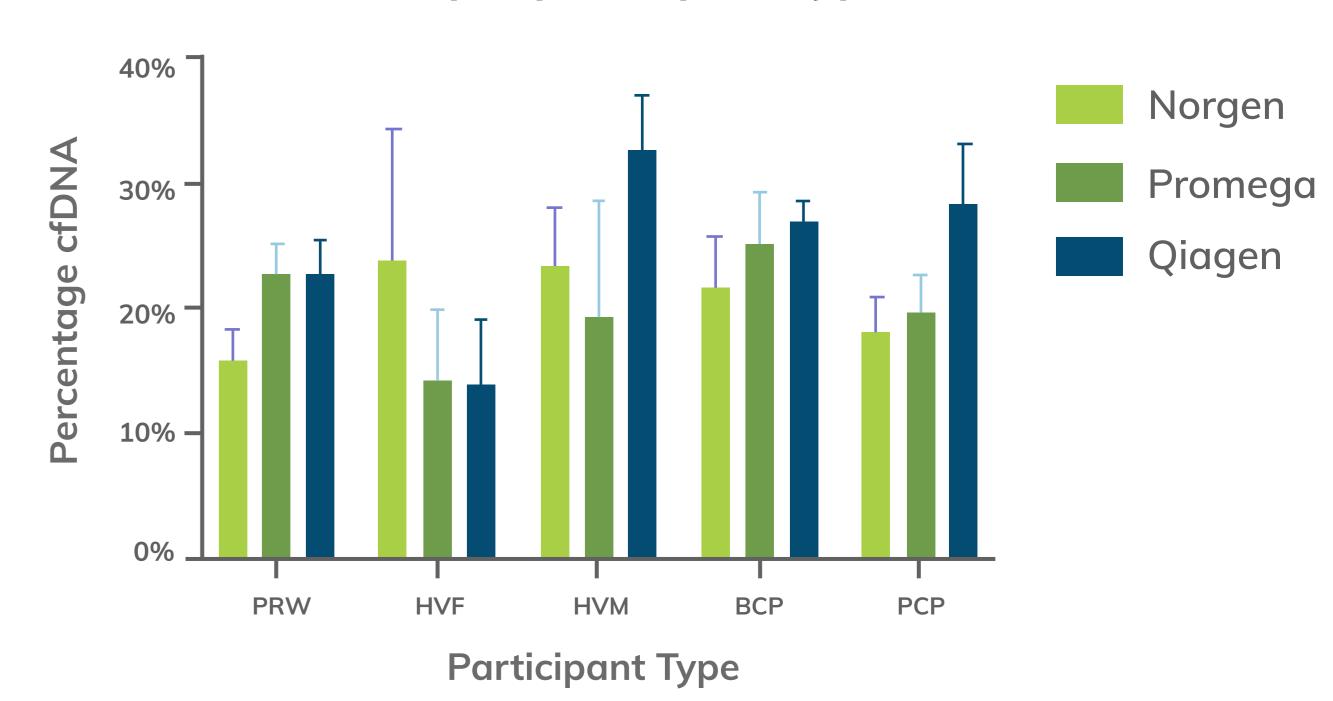


Fig 3. Comparison isolation methods based on cfDNA percentage per participant type

Depicted as mean ± SEM, BCP: breast cancer patients; HVF: healthy female volunteers; HVM: healthy male volunteers; PCP: prostate cancer patients; PRW: pregnant women

Almost all participants rated the usability of Colli-Pee[®] Large Volumes, before, during and after collection high (80 on scale of 100). In addition, about 80% of the participants rated the general usability of Colli-Pee[®] Large Volumes as easy to very easy. And 71% preferred Colli-Pee[®] over a regular urine cup (29%) for the collection of urine.

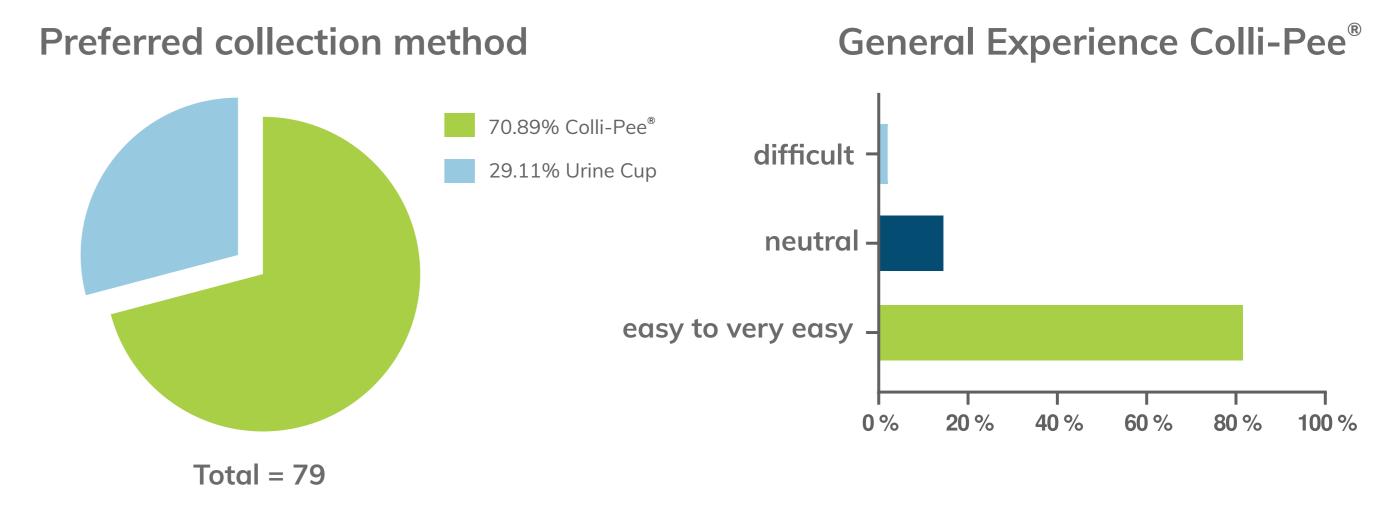


Figure 4: Usability results summary

CONCLUSION

Performance of all three commercially available cfDNA isolation kits was comparable. Also, Colli-Pee[®] Large Volumes showed good usability results.





