Application Note

QIAxpert® — a powerful system for nucleic acid quality control

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Introduction

Determining the purity and concentration of nucleic acid samples is an essential application in molecular biology today, especially for complex workflows. One of the greatest challenges is ensuring that each nucleic acid sample has an adequate concentration and that any chemical contaminants or other impurities are detected. Several analysis systems are available for this purpose, such as the QIAxpert (QIAGEN GmbH), NanoDrop® (Thermo Fisher Scientific), Qubit® Fluorimeter (Thermo Fisher Scientific), DropSense® 96 (Trinean), and DS-11 Spectrophotometer (DeNovix). We compare the accuracy and sensitivity of these systems, and demonstrate that QIAxpert has a unique combination of features that make it the most valuable option for nucleic acid quality control.

Materials and methods

We used calf thymus DNA purified with QIAamp® chemistry on the QIAcube® and commercial human reference RNA to compare the linearity among the different systems. We set up a serial dilution of the DNA and RNA samples and measured 5 replicates of each dilution step on the QIAxpert system (DNA QIAamp or RNA RNeasy application, respectively), the Nanodrop 8000 and the Qubit.

In addition, we measured and analyzed concentrations of 180 genomic DNA and 105 RNA samples. Measurements were performed in triplicate on the DeNovix® DS-11 Spectrophotometer, the Trinean DropSense 96 and the QIAxpert system (DNA QIAamp or RNA RNeasy application, respectively). The analyzed genomic DNA and RNA samples had different concentrations and had been stored at –20°C over different time periods. Thus, the validation setup represents the wide range of samples collected and analyzed by researchers. Measurements were made in batches of 16 DNA and RNA samples.

Results

Early detection of sample contaminants is crucial in workflows using nucleic acids. Because specific spectral profiling gives insight into the quality and composition of analyzed samples, we initiated our studies with spectrophotometric measurement of microvolume DNA samples using QIAxpert. We compared pure calf thymus DNA purified using QIAamp chemistry (Figure 1A) and purified calf thymus DNA spiked with RNA (Figure 1B). The spectral content profiles, presented in Figure 1, reveal the exact contents of the samples and demonstrate the accuracy and ease with which DNA and RNA can be distinguished. QIAxpert is a reliable and fast tool for discriminating between nucleic acids and detecting contaminants without the need for dyes or time-consuming preparation steps.

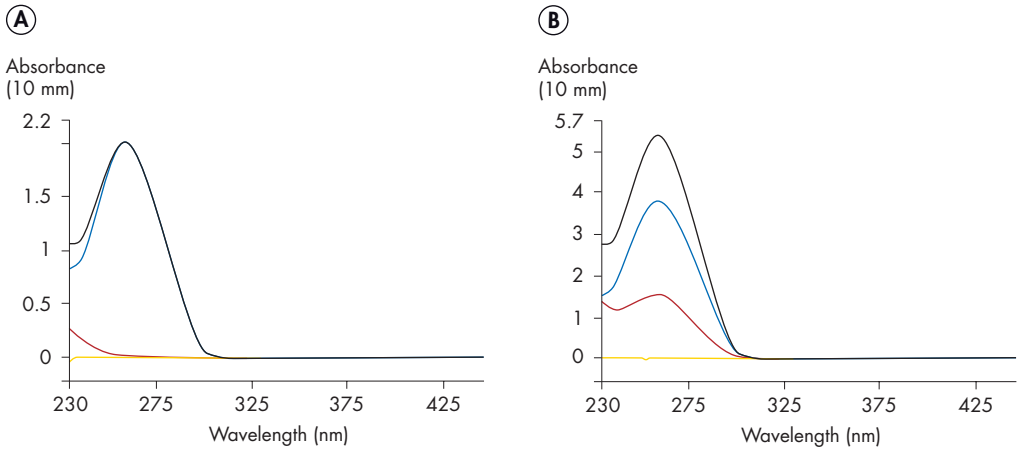

Figure 1. Spectral content profiling discriminates between components in complex samples. **A:** A pure sample of calf thymus DNA shows no significant contamination, as evidenced by the simple spectrum. **B:** A sample of DNA spiked with RNA was accurately analyzed and quantified. Both samples were measured on the QIAxpert DNA QIAamp application. DNA is indicated by the blue absorbance line while RNA and all detected impurities are depicted by the orange line. A gray line typically appears as a result of the sample background spectrum. Due to low sample background in this case, the gray line is not visible. A yellow line typically depicts the residual spectrum that cannot be attributed to reference profiles used in the algorithm. In this case, the yellow line is flat because no residual components could be detected. The sum of nucleic acid content, impurity and residual spectrum is represented by the black line.

To test the linearity of the system, we prepared samples of various concentrations ranging from 1 to 2000 ng/µl from a commercial solution of pure salmon sperm DNA. Each dilution was measured 5 times using 2 µl samples pipetted onto QIAxpert slides. The results demonstrate high concordance of the quantifications made on the QIAxpert system with the target concentrations over the complete range of concentrations measured (Figure 2).

Subsequently, we compared the measurement linearity of QIAxpert with two other systems, Nanodrop and Qubit. The performance of QIAxpert is demonstrably more reliable than that of Nanodrop and Qubit. Both systems consistently misestimated DNA and RNA sample concentrations. The DNA and RNA concentrations reported by Nanodrop were consistently overestimated (Figure 3, top and bottom middle graphs), while Qubit consistently underestimated DNA concentration values (Figure 3, top right graph) and overestimated RNA concentration values (Figure 3, bottom right graph). In contrast, the measurements from QIAxpert were most accurate, matching the true concentrations of the analyzed samples (Figure 3, top and bottom left graphs).

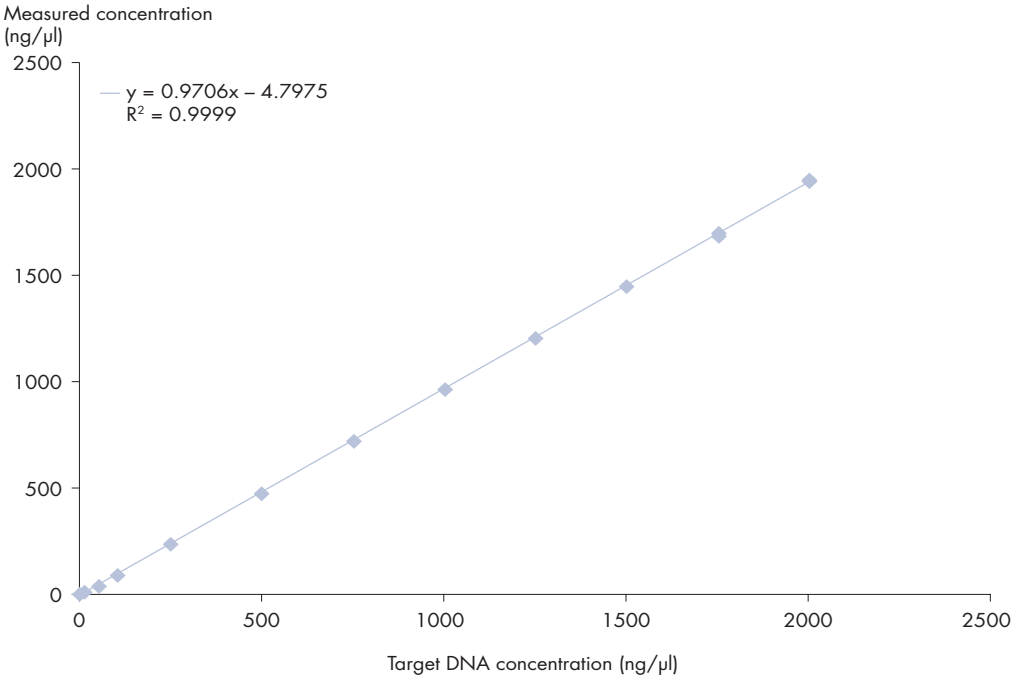

Figure 2. Excellent measurement linearity of QIAxpert. The spectrophotometry data were plotted against the target concentration. The data demonstrate outstanding linearity for samples with concentrations up to 2000 ng/µl or 40 OD (A_{260}, 10 mm), respectively. The calculated regression coefficient is >0.999.

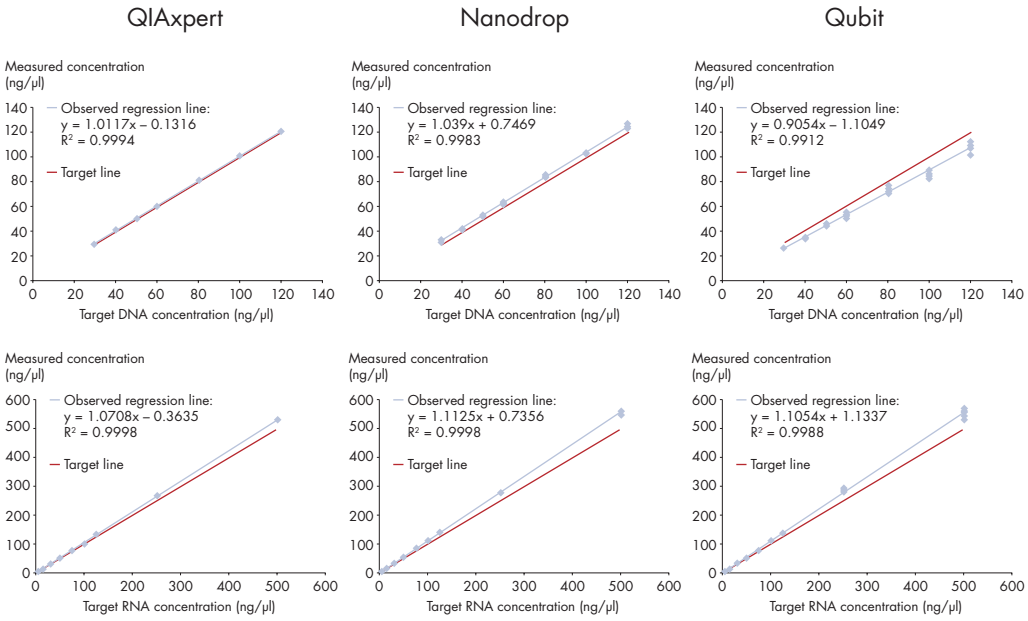

Figure 3. Superior linearity of concentration measurements for commercial DNA and RNA samples using QIAxpert. Top row: A dilution series of calf thymus DNA measured with the QIAxpert, Nanodrop 8000 and Qubit systems. Data shown for QIAxpert reflect total nucleic acid concentrations measured with the DNA QIAamp app. Bottom row: A dilution series of human reference RNA measured with the QIAxpert, Nanodrop 8000 and Qubit systems. Data shown for QIAxpert reflect total nucleic acid concentrations measured with the RNA RNeasy® app.

In addition, field samples of genomic DNA and total RNA were analyzed by the Human Genomics research group in their laboratories at the Department of Biomedicine, University Hospital Basel, to compare QIAxpert, DropSense 96 and the DS-11 Spectrophotometer. All concentration measurements were comparable among the three systems (Figure 4). Some samples exhibited visible measurement deviation in one system compared to the others. This was due to the varying sample qualities in terms of viscosity, degree of buffer evaporation, and contamination. Overall, however, QIAxpert proved to be at least as reliable as the other two tested systems for the measurement of field samples.

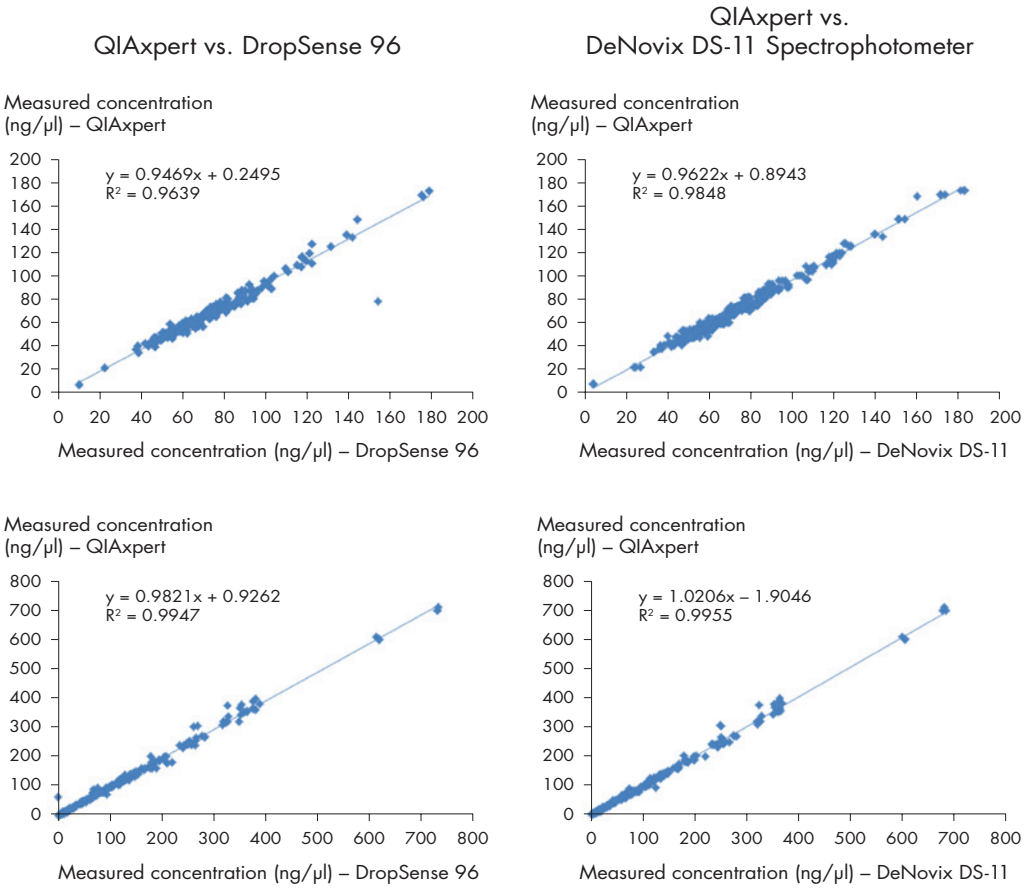

Figure 4. Reliable measurement performance of QIAxpert for field DNA and RNA samples. QIAxpert, DropSense 96 and the DeNovix DS-11 Spectrophotometer were employed to measure concentration of field DNA and RNA samples. Right: Analyses of the DNA (top) and RNA (bottom) samples with the DeNovix DS-11 Spectrophotometer and QIAxpert yielded highly comparable results, indicated by the regression line slopes being close to 1. Left: Similarly, results with the DropSense 96 were comparable.

QIAxpert can analyze up to 16 samples in less than 2 minutes and enables rapid discrimination between sample components, contaminant detection and accurate concentration measurements of microvolumes of nucleic acids. QIAxpert also offers more flexibility in terms of data reporting. Results are easily exported to a USB stick or a smart device. QIAxpert thus offers a unique combination of features that make it an attractive alternative to other nucleic acid analysis systems on the market.

Conclusions

In conclusion, QIAxpert:

- Differentiates between DNA and RNA within a single sample measurement
- Accurately identifies contaminants after purification with selected QIAGEN kits
- Uses dedicated slides that prevent sample cross-contamination

Compared to the other systems in this evaluation, the QIAxpert system is a more reliable, flexible and affordable system for nucleic acid quality control.

References

1. Schade, C. (2014) Quality Control: An important success factor in nucleic acid-based analysis. American Laboratory 46.
2. Schade, C. (2014) Kontaminationsdetektion, Qualitätskontrolle von Nukleinsäuren. BIOspektrum 02/14, 182.

Ordering Information

Product	Contents	Cat. no.
QIAxpert Instrument	QIAxpert instrument, including 1 year warranty on parts and labor	9002340
QIAxpert Slides 40	25 disposable microfluidic slides for analyzing up to 16 samples per run	990700
QIAxpert System	QIAxpert instrument with installation and startup training and 1 year warranty coverage including parts, labor and shipping. Repair by sending to regional repair center.	9002368
QIAxpert System Full 2 Year Package	QIAxpert instrument with installation, startup training and 2 years full warranty coverage*	9002364
QIAxpert System Full 3 Year Package	QIAxpert instrument with installation, startup training and 3 years full warranty coverage*	9002367

* Full warranty coverage: coverage of parts, labor, shipping; repair by sending to a regional repair center; loaner system provided within 2–3 working days. Please contact your local sales representative for other service options.

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