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Technical Note
PAXgene® Blood miRNA System

Comparison of miRNA recovery from the PAXgene Blood miRNA System and the Tempus™ Blood RNA Tube processed with the Tempus Spin RNA Isolation Kit

Study Design

Whole blood was collected into one PAXgene Blood RNA Tube* and one Tempus Blood RNA Tube† from each of six apparently healthy, consented adult subjects. Both PAXgene and Tempus tubes were stored for one day at room temperature prior to RNA extraction. RNA from the PAXgene tubes was extracted using the PAXgene Blood miRNA Kit†. RNA from the Tempus tubes was extracted using the Tempus Spin RNA Isolation Kit†, including the optional DNase treatment.

Three representative miRNAs, mir-16, mir-30b, and mir-103, were quantified using qRT-PCR performed on an ABI PRISM® TaqMan® 7900 System† with the QuantiTect® SYBR® Green RT-PCR Kit†. The miRNA assays were performed in triplicate (data on file at QIAGEN GmbH, Hilden, Germany).

Results

Figure 1 depicts cycle threshold‡ (C_T) values for three different miRNAs isolated from blood collected from six subjects in either PAXgene or Tempus tubes as measured by qRT-PCR. Table 1 describes the summary statistics for Figure 1.

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* IVD CE-marked / 510(k) cleared (k042613).

† For research use only. Not for use in diagnostic procedures. No claim or representation is intended to provide information for the diagnosis, prevention, or treatment of a disease.

‡ C_T is defined as the threshold cycle number in which a fluorescent signal is detected above background. A C_T value is the number of cycles needed for the fluorescence signal to exceed the background. There is, therefore, an inverse relationship between C_T and miRNA concentration: the lower the C_T number, the higher the miRNA concentration in purified RNA eluates.

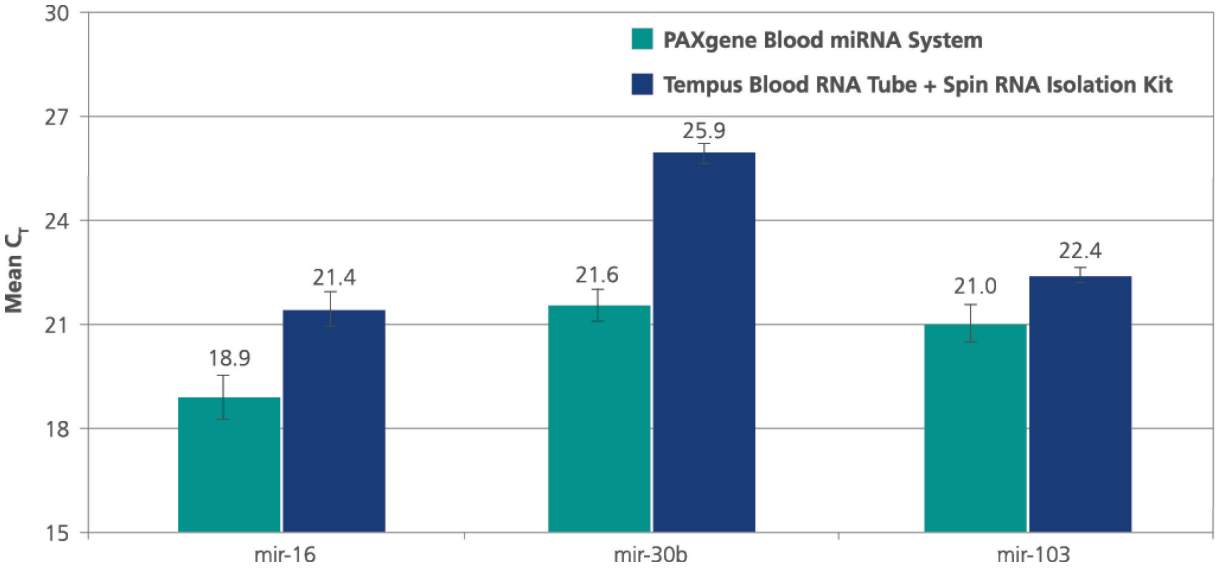

Figure 1. Mean C_T values for RT-PCR assays (run in triplicate) of three miRNAs isolated from blood collected into either PAXgene or Tempus Blood RNA Tubes. The miRNA species mir-16, mir-30b, and mir-103 were amplified from RNA eluates generated with the PAXgene Blood miRNA System or the Tempus Blood RNA Tubes and Tempus Spin RNA Isolation Kit. Numbers indicate the mean C_T value for each miRNA-tube pair.

Table 1. Summary statistics for the amplification of three miRNAs

Statistic	C_T Value by miRNA and collection tube					
	mir-16		mir-30b		mir-103	
	PAXgene	Tempus	PAXgene	Tempus	PAXgene	Tempus
Min	17.7	20.4	20.7	25.5	20.0	21.0
Max	19.9	21.9	22.3	26.5	21.7	22.7
Mean	18.9	21.4	21.6	25.9	21.0	22.4

As illustrated in Figure 1, mean C_T values for all three miRNAs were lower in RNA eluates purified from blood collected in PAXgene Blood RNA Tubes than those for blood collected in Tempus Blood RNA Tubes. Specifically, compared to RNA eluates from Tempus Blood RNA Tubes, these differences were −2.5 C_T for mir-16, −4.3 C_T for mir-30b, and −1.4 C_T for mir-103. As higher C_T values correspond to fewer target sequences, the lower C_T values indicate that the PAXgene Blood miRNA System yields 3 to 20-fold more miRNA from PAXgene tubes for mir-16, mir-30b, and mir-103, compared to Tempus Blood RNA Tubes.

Conclusion

For all miRNA species examined in this study, the PAXgene Blood miRNA System, comprising the PAXgene Blood RNA Tube and the PAXgene Blood miRNA Kit optimized for enrichment of small RNA, recovered 3–20 fold higher amounts of miRNA from whole blood than the combination of the Tempus Blood RNA Tube and the Tempus RNA Spin Kit.

Products used:

Product	Catalog No.
PAXgene Blood RNA Tubes (100)	762165
PAXgene Blood miRNA Kit (50)	763134
QuantiTect SYBR Green RT-PCR Kit (200) (QIAGEN)	204243

For up-to-date licensing information and product-specific disclaimers, see the respective PreAnalytiX® kit handbook or user manual. PreAnalytiX kit handbooks and user manuals are available at www.qiagen.com or can be requested from QIAGEN Technical Services or your local distributor.

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