Comparison of different brands of PCR tubes

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PCR tubes with thinner side walls have faster heating and cooling kinetics. We recommend thin-wall tubes from Sorenson BioScience (Cat. No. 16950, VWR Cat. No. 53550-106) and DiaMed (Cat. No. AD0210-FCN).

Introduction

Batched real-time PCR machines such as the LightCycler® (Roche) and SmartCycler® (Cepheid) require the use of proprietary tubes. Other batched machines such as the Rotor-Gene™ (Corbett Life Science) use industry-standard 0.2 ml PCR tubes. The Spartan DX™ is a non-batched personal DNA analyzer that also uses industry-standard tubes.

Industry-standard PCR tubes from various manufacturers are typically made of polypropylene plastic and have roughly the same dimensions. That being said, there are variations in their exact dimensions and side wall thicknesses. PCR is affected by the efficiency of heat transfer from a heat source to the reaction mixutre (Ref 1). It would be expected that tubes with thinner side walls would conduct heat faster than tubes with thicker side walls.

The purpose of this study was to determine the dimensions, temperature robustness, and heating and cooling kinetics of four different brands of PCR tubes (Axygen, Light Labs, Sorenson BioScience, and DiaMed) with the Spartan DX.

Materials and Methods

Determination of PCR tube dimensions

Dimensions of four different brands of 0.2 ml PCR tubes were measured by digital caliper (ULTI 83071). The brands examined included Axygen (Cat. No. PCR 02C), Light Labs (Cat. No. A-1001), DiaMed (Cat. No. AD0210-FCN), and Sorenson Bioscience (Cat. No. 16950, VWR Cat. No. 53550-106).

Temperature robustness

The four brands of tubes were heated at 110°C for 20 min followed by an examination for visual signs of warping or deformation.

Determination of liquid temperatures

A thermocouple (Omega, Part# 5SRTC-TT-T-40-72) was used to measure the reaction liquid temperatures achieved with the cycling parameters listed in Table 1. Specifically, the lids of the four different PCR tubes were altered in the following manner: a hole 1 mm in diameter was drilled into the top of the tube, the thermocouple was threaded through the hole, and the tip of the thermocouple was positioned 1-2 mm from the bottom and equidistant from the sides. The tubes were then filled with 20 µl of distilled water, and topped with 15 µl of mineral oil. The modified lid was then used to cap the tubes and temperature data from the thermocouple was logged with a Data Logger Thermometer (Fluke 54II) and analyzed using FlukeView Forms software (Ref 2). The temperature was monitored every second over the duration of the cycling parameters and high and low values were recorded.

Results

Table 2 shows that the average length of the tubes ranged from 20.81 to 20.93 mm, the diameter ranged from 5.95 to 6.07 mm, and the thickness ranged from 0.32 to 0.41 mm. In

Step	Temperature	Time	Cycles
Initial denaturation	93.8°C	30 s	1
Denaturation	93.8°C	30 s	5
Annealing/extension	57.1°C	30 s	5

Table 1. Cycling parameters.

Company	Length (mm ± S.D.)	Diameter (mm ± S.D.)	Thickness (mm ± S.D.)
Axygen	20.81 ± 0.07	6.07 ± 0.03	0.41 ± 0.03
Light Labs	20.93 ± 0.08	5.99 ± 0.04	0.39 ± 0.07
Sorenson	20.91 ± 0.04	5.95 ± 0.04	0.32 ± 0.04
DiaMed	20.86 ± 0.04	6.00 ± 0.04	0.32 ± 0.01

Table 2. Average dimensions of PCR tubes (n=10).

Company	Liquid Temperatures (°C)	
	Denaturation	Annealing/Extension
Axygen	81.5	62.8
Light Labs	83.5	61.5
Sorenson	84.6	59.0
DiaMed	83.7	60.1

Table 3. Average liquid temperatures achieved over five cycles.

terms of temperature robustness, none of the tubes showed any deformation after 20 min at 110°C. Table 3 shows that thinner-walled tubes reached higher denaturation temperatures and lower annealing/extension temperatures. Specifically, the Axygen tube had the thickest side wall and reached denaturation temperatures that were 2.0 to 3.1°C lower than tubes from Light Labs, DiaMed, and Sorenson Bioscience. Similarly, the Axygen tube achieved annealing/extension temperatures that were 1.3 to 3.8°C higher than the other tubes. For certain primers, it was found that the Axygen tube's slower rates of heating and cooling resulted in significant delays to threshold cycle values for real-time PCR (data not shown).

Discussion and Conclusions

This study found that PCR tubes with thinner walls had faster heating and cooling kinetics compared to tubes with thicker walls. The most likely explanation is that plastic is an insulator and heat is transferred more efficiently through thinner plastic.

In conclusion, we recommend 0.2 ml flat cap PCR tubes from Sorenson Bioscience (Cat. No. 16950, VWR Cat. No. 53550-106) or DiaMed (Cat. No. AD0210-FCN) for use with the Spartan DX.

References

1. Schoder D et al. (2005). Novel approach for assessing performance of PCR cyclers used for diagnostic testing. *Journal of Clinical Microbiology*. 43(6):2724-2728.

2. Liu R et al. (2007). Effect of increasing reaction volumes on PCR. *Spartan Bioscience*. AN 008: 1-2.