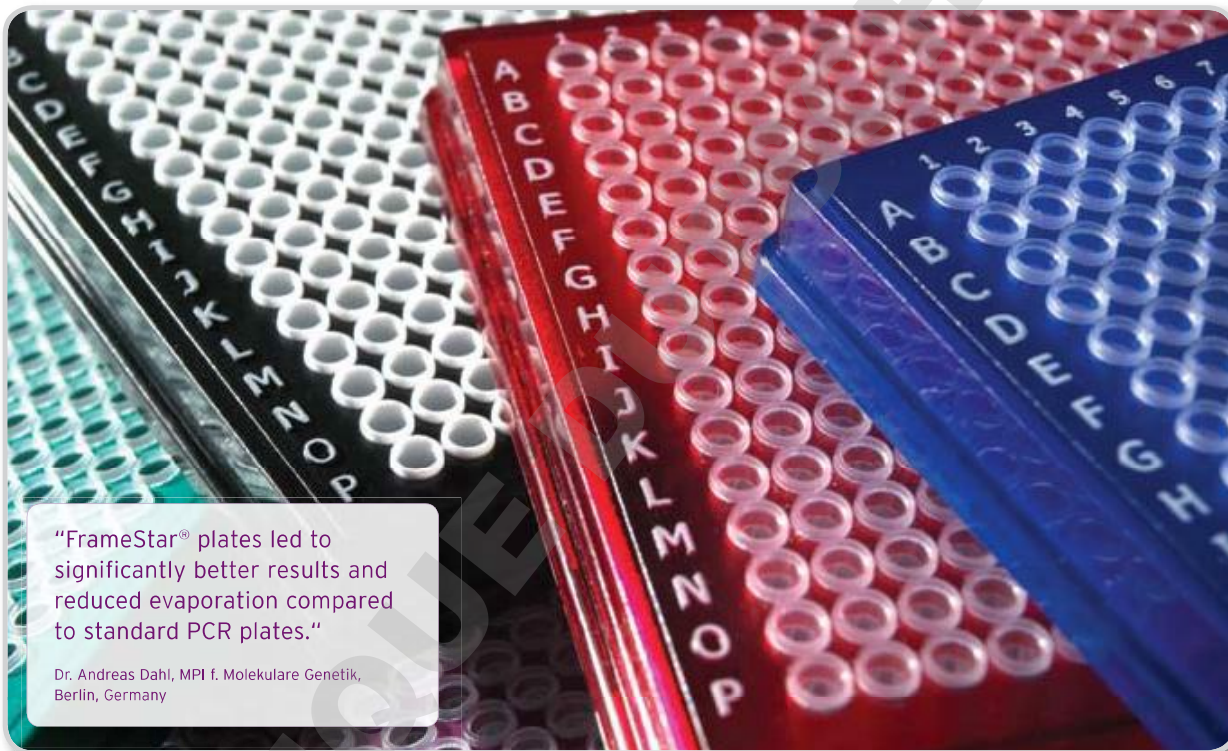


FRAMEstar® Attitude®

THE LOW VOLUME PCR PLATE

FrameStar® two-component technology reduces evaporation from PCR plates, improving results and allowing for volume reductions to save money.



"FrameStar® plates led to significantly better results and reduced evaporation compared to standard PCR plates."

Dr. Andreas Dahl, MPI f. Molekulare Genetik,
Berlin, Germany

FrameStar® PCR plates maximise thermal stability at high temperatures which prevents sample loss by minimising thermal expansion during PCR. The two-component design combines the advantages of thin wall polypropylene tubes for optimum PCR results and a rigid polycarbonate skirt and deck for highest thermal stability and rigidity. In contrast to standard one piece PCR plates evaporation from corner positions and outside rows of wells is minimal which allows for downscaling of reagent volumes and saves costly reagents.

- Improved PCR results
- Minimised sample loss
- No increase of sample evaporation from corner wells
- Recommended for low volume PCR
- Thin-wall design for fast PCR
- Cost saving through downscaling of reaction volumes
- Ideal for robotics, as plate distortion eliminated post PCR

FrameStar PCR plates are covered by one or more of the following U.S. patents or their foreign counterparts, owned by Eppendorf AG: US Patent Nos. 7,347,977 and 6,340,589

Thermal expansion of Polypropylene (PP) plates leads to evaporation from outside wells.

PP is the optimum material for PCR tubes. It provides the most efficient heat transfer, as well as an inert surface with low binding capabilities for nucleic acids, proteins and other molecules. However, the material is not thermally stable in a plate format and expands and contracts during each PCR cycle (Figure 1). Such thermal expansion will weaken the plate seal and leads to sample evaporation mainly from corner wells and outside rows.

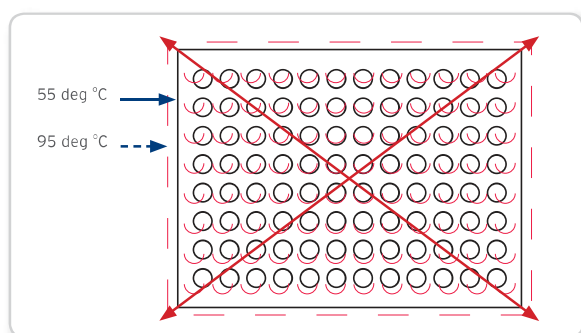


Figure 1: Standard plates with polypropylene frame expand by up to 2mm during thermal cycling which leads to movement of wells away from the plate centre. This movement is most significant in corner positions and outside rows of the plate. Sealing sheets do not expand at this rate so that the movement of wells will weaken the seal and lead to evaporation especially in corner positions and outside rows.

Thermal cycler Blocks do not prevent thermal expansion of PCR plates.

PCR blocks do not support PCR plates from the sides and the high temperatures from the thermal block and heated lid accelerate expansion of the plates (Figure 2).

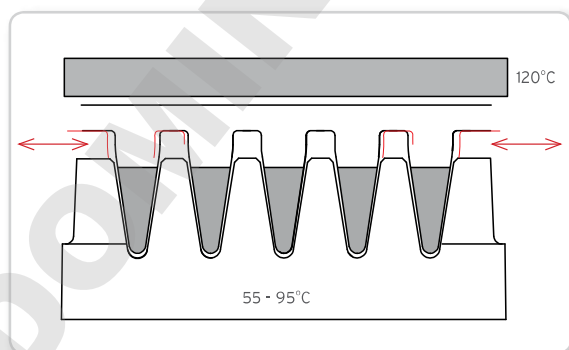


Figure 2: Side-on view of a PCR plate in a thermal cycler. The sealed plate is sandwiched between the cycler block and the heated lid but it is only partly fixed in position at the bottom of tubes, allowing the plate to expand horizontally.

FrameStar® 2-component technology reduces thermal expansion and sample evaporation

The polycarbonate frame of FrameStar® plates is more heat resistant than standard PP plates which reduces thermal expansion to a minimum. For this reason the seal integrity remains intact even at elevated temperatures during PCR.

To illustrate this advantage of our two-component technology we have compared evaporation from one piece PP plates and FrameStar® PCR plates: Each well of a non-skirted 96well plate (single piece, PP) and a Framestar® non-skirted design (code 4ti-0710) was filled with 10µl H₂O. The plates were sealed with a QPCR adhesive (code 4ti-0560) and the weight of plates was measured before and after performing PCR (30 cycles x 15" 95°C; 15" 55°C).

Table 1 shows that the average volume loss from one piece PP plates was 2.3µl per well which is equal to 23% of the total reaction volume. In contrast the volume loss from FrameStar plates was only 0.49µl per well.

Plate Type	Starting weight	Weight post PCR	Weight loss	Volume loss total/per well
Framestar 4ti-0710	26.678g	26.631g	0.047g	47µl/0.49µl
One piece	17.807g	17.586g	0.221g	221µl/2.3µl

Table 1: Weight and volume loss from 96well PCR plates. Results shown are averages from 5 plates of each plate type. One piece PP plates showed more than 4 times higher volume loss than FrameStar® plates.

"Reducing the SyBr Green consumption by 40% means considerable cost savings to us."

Carolin Deiner, PhD

"For our low volume real-time PCR assays the optimal combination we found were the 4titude® two component "FrameStar®" 384well PCR plates in combination with heat sealing."

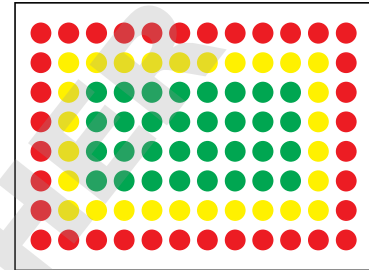
Dr. Andreas Dahl, MPI f. Molekulare Genetic, Berlin, Germany

FrameStar® PCR plates minimise sample loss across the plate

Evaporation from one piece PP plates is highest in the outer wells

Since thermal expansion and movement of wells in one piece PP plates is enhanced around the edges of the plates (see Figure 1) evaporation is the highest from the two outer rows of wells. Figure 3 illustrates the levels of sample evaporation from different areas of PP plates. Only the inner 32 wells of a one piece 96well plate show low levels of evaporation but sample loss is high from the two outside rows which contain more than 65% of the wells.

Figure 3: Evaporation from the outside rows (red) is highest, medium level evaporation occurs in the second row (yellow) and sample loss from the inner 32 wells is lowest.



FrameStar® 2-component plates improve consistency of PCR results

We have compared the degree of evaporation from different areas of one piece PP and FrameStar® PCR plates. First, the 64 outer wells (two outside rows) of both plate types were filled with 10µl H₂O, plates were then sealed with a QPCR adhesive (code 4ti-0560) and their total weight determined before and after PCR. The experiment was repeated with a set

of plates of which the inner 32 wells (green area in Figure 3) were filled. Table 2 shows that evaporation from outer wells of standard PP plates was significant and 65% higher than from inner wells. In contrast, FrameStar plates showed low levels of sample loss across the plate.

Weight and Volume loss

Plate Type	Starting weight	Weight post PCR	Weight loss	Volume loss total/per well
Framestar 4ti-0710 outer 64 wells	26.230g	26.193g	0.037g	37µl/0.57µl
One piece, outer 64 wells	17.299g	17.118g	0.181g	181µl/2.8µl
Framestar 4ti-0710 inner 32 wells	25.841g	25.824	0.017g	17µl/0.53µl
One piece, inner 32 wells	17.132	17.078	0.054g	54µl/1.69µl

Table 2: Weight and volume loss from different sections of 96well PCR plates. Results shown are averages from 5 plates of each plate type. Volume loss from the outside wells of one piece PP plates was 5-times higher than from FrameStar® plates.

The above results show that reaction volumes remain consistent across the 96wells (or 384wells, data not shown) in FrameStar® plates. In contrast, the reaction volumes in standard plates will differ increasingly between wells during

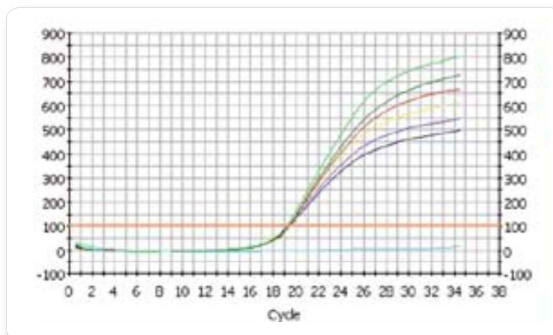
PCR. Buffer concentrations in outside rows will increase dramatically and result in reduced enzyme activity. In extreme cases samples will fully evaporate.

FrameStar® 2-component technology allows for reduction of assay volumes and cost

Due to the much improved seal integrity, reaction volumes can often be reduced when using FrameStar® plates. Such downscaling of experiments can be successfully implemented

without any loss of assay sensitivity or consistency (Figure 4) and reagent savings can be considerable.

Figure 4: Comparable Ct Values and mean deviations were obtained with 15µl and 25µl SyBr Green assays in heat sealed FrameStar® plates.



	Threshold Cycle Ct	Identifier
C4	19.4	ubi 15-5µl templ
C5	19.3	ubi 15-5µl templ
C6	19.1	ubi 15-5µl templ
C7	19.4	ubi 25-5µl templ
C8	19.2	ubi 25-5µl templ
C9	19.4	ubi 25-5µl templ
E6	N/A	neg ubi- 5µl

FRAMEstar®

PRODUCT RANGE



FrameStar 384:
384well plate, ideal for robotic use.
Plate code A



FrameStar 480/384:
384well, designed for the Roche LightCycler® 480.
Plate Code B



FrameStar 96 skirted low profile:
96well plate, low profile, recommended for robotics and low volume PCR.
Plate Code C



FrameStar 480/96 semi-skirted:
96well plate, low profile, designed for the Roche LightCycler® 480.
Plate Code D



FrameStar FastPlate 96 semi-skirted low profile:
96well plate, low profile, recommended for ABI Fast Block cyclers.
Plate Code E



FrameStar 96 with upstand:
96well semi-skirted plate with upstand, standard profile, designed to fit ABI thermal cyclers and sequencers.
Plate Code F



FrameStar 96 semi-skirted (cut corner A12):
96well semi-skirted plate, standard profile, A12 cut corner to fit ABI instruments.
Plate Code G



FrameStar 96 semi-skirted (cut corner H1):
96well semi-skirted plate, standard profile, cut corner H1, designed for universal fit.
Plate Code H



FrameStar 96 non-skirted:
96well plate, standard profile, fits most thermal cyclers.
Plate Code I



FrameStar 96 non-skirted low profile:
96well plate, low profile, recommended for low volume PCR.
Plate Code J

Ordering Information

Choose plate type from selection above and obtain Plate Code.
Choose colour combination required and obtain Part Number.

All pack sizes are 50 plates per case

Plate Code (from above)	Purple Frame	Blue Frame	Clear Frame	Green Frame	Red Frame	Black Frame	Black Frame	Clear Frame
	Clear Wells	Clear Wells	Clear Wells	Clear Wells	Clear Wells	Clear Wells	White Wells	White Wells
A	4ti-0384	4ti-0384/B	4ti-0384/C	4ti-0384/G	4ti-0384/R	4ti-0384/X	4ti-0385	
B			4ti-0380/C					4ti-0381
C	4ti-0960	4ti-0960/B	4ti-0960/C	4ti-0960/G	4ti-0960/R	4ti-0960/X	4ti-0961	
D			4ti-0950/C					4ti-0951
E			4ti-0910/C					4ti-0911
F	4ti-0730	4ti-0730/B	4ti-0730/C	4ti-0730/G	4ti-0730/R	4ti-0730/X	4ti-0731	
G			4ti-0770/C				4ti-0771	
H			4ti-0900/C					4ti-0901
I	4ti-0710	4ti-0710/B	4ti-0710/C	4ti-0710/G	4ti-0710/R	4ti-0710/X	4ti-0711	
J	4ti-0720	4ti-0720/B	4ti-0720/C	4ti-0720/G	4ti-0720/R	4ti-0720/X	4ti-0721	

See www.4ti.co.uk/technical/compatibility.php for compatibility with thermal cyclers & sequencers

All skirted plates are available with linear barcodes. Please see www.4ti.co.uk/technical/barcode.php for further details and a custom order form.

The North Barn, Surrey Hills Business Park, Damphurst Lane,
Wotton, Surrey RH5 6QT, UK

Tel: +44 (0)1306 884885 Fax: +44 (0)1306 884886

Email: sales@4ti.co.uk Web: www.4ti.co.uk