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ProductInformation

PCR Low Ladder Marker Set

Product Code D 7808

Store at 2 to 8 °C

Product Description

The PCR Low Ladder Marker Set has been especially designed for size determination of PCR generated DNA fragments. Mixing the two markers together will produce a band every 20 bp, with every fifth band (100 bp increments) more intense. These markers can be used with either agarose or polyacrylamide gels.

Fragment Sizes: base pairs (bp)

20 bp Ladder				100 bp Ladder
1,000	700	400	100	1,000
980	680	380	80	900
960	660	360	60	800
940	640	340	40	700
920	620	320	20	600
900	600	300		500
880	580	280		400
860	560	260		300
840	540	240		200
820	520	220		100
800	500	200		
780	480	180		
760	460	160		
740	440	140		
720	420	120		

Components

Suitable for 50 applications of the 20 bp ladder and approx. 150 applications of the 100 bp ladder

- 25 µg vial of 20 bp ladder, Product Code P 2849, containing 50 bands ranging from 20 to 1,000 bp in exact 20 bp increments. (Approx. 200 µg/ml)
- 25 µg vial of 100 bp ladder, Product Code P 9364, containing 10 bands ranging from 100 to 1,000 bp in exact 100 bp increments. (Approx. 100 µg/ml)
- 1 ml vial of 6X Sample Loading Buffer, Product Code B 6924, containing 15% (w/v) Ficoll, 0.06% (w/v) bromophenol blue, 0.06% (w/v) xylene cyanol FF, 30 mM EDTA.

Storage/Stability

Markers are stable for 12 months when stored at 2-8 °C. This product is shipped at ambient temperature. Routinely used marker should be stored at 2-8 °C. For infrequently used markers, storage at –20 °C may increase shelf life.

Storage Buffer for Ladder Components 10 mM Tris-HCl, pH 7.5-8.0

1.0 mM EDTA

Product Summary

Suitable for use as electrophoresis marker for DNA. Typically, 1.5-2.0 μ l of DNA ladder component, 2.5 μ l of 6X loading buffer plus 10.5 μ l of water is used. This translates to 15-20 ng/band, giving sharp, easily visible and photographed bands. Adjustments may be made for different well sizes and individual preferences.

Suitability Assay

Each DNA Ladder was prepared for electrophoresis as follows:

20 bp DNA Ladder

2.5 µl 20 bp DNA Ladder

5.8 ul water

1.7 µl 6X Loading Buffer (Product Code B 6924)

100 bp DNA ladder

1.6 µl 100 bp DNA Ladder

6.7 ul water

1.7 µl 6X Loading Buffer (Product No. B 6924)

10 μ l of each of the above solutions were loaded on a 20 cm, 3% agarose gel (agarose, wide range:routine, 3:1, Product Code A 7431). The gel was run with the appropriate DNA standards at 4 °C in 1X TBE (Product Code T 9525) for approximately 16 hours, until the bromophenol blue tracking dye reaches the bottom of the gel. **Note:** The 20 bp band of the 20 bp ladder may run off the end of the gel. After staining for 15-20 minutes in 5 μ g/ml ethidium bromide and destaining with water for 15-20 minutes, the resulting banding pattern for each ladder was consistent with the indicated DNA sizes.

<u>Factors Influencing Resolution and Accurate Sizing of DNA Fragments</u>

- DNA loading: For accurate size determinations, load the smallest practical amount of sample DNA. A single application of 10-20 ng is readily visible and will yield a sharp, accurately sizable band.
- 2. Well thickness: For best resolution of DNA bands, use only properly formed sample wells, ≤1 mm in thickness.
- 3. Salt concentration: It is important to accurately match the salt concentration of the ladder mixture to that of the DNA being evaluated in order to obtain the best size determinations. One useful technique for very precise sizing of sample-fragments, which eliminates concerns over matching salt concentrations, is to co-electrophorese the sample and the ladder in the same well. Ladder-only and sample-only lanes should be run to aid in interpretation of electrophoresis patterns.
- Anomalies: The 100 bp ladder may show a doubleor triple-banding pattern in some types of polyacrylamide gels, particularly under higher run temperatures.

7/02