ZYMO RESEARCH

The Beauty of Science is to Make Things Simple

INSTRUCTION MANUAL

Genomic DNA Clean & Concentrator™-10

Catalog Nos. **D4010 & D4011**

Highlights

- Quick (5 minute) spin column recovery of large-sized DNA (e.g., genomic, mitochondrial, plasmid (BAC/PAC), viral, phage, (wga)DNA, etc.) from any enzymatic reaction or impure preparation (e.g., Proteinase K digestion). *No messy precipitations!*
- Unique spin column for low volume (≥10 µl) elution of ultra-pure, high-yield DNA.
- Eluted DNA is ideal for PCR, restriction endonuclease digestion, Sanger and Next-Gen sequencing, etc.

Contents

For Research Use Only

Ver. 1.0.0

ZYMO RESEARCH CORP.
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Satisfaction of all Zymo Research products is guaranteed. If you should be dissatisfied with this product please call 1-888-882-9682.

Product Contents

Genomic DNA Clean & Concentrator™-10 (Kit Size)	**D4010** (25 Preps.)	**D4011** (100 Preps.)	**Storage Temperature**
ChIP DNA Binding Buffer	50 ml	2 x 50 ml	Room Temp.
DNA Wash Buffer[1]	6 ml	24 ml	Room Temp.
DNA Elution Buffer	1 ml	4 ml	Room Temp.
Zymo-Spin™ IC-XL Columns	25	100	Room Temp.
Collection Tubes	50	100	Room Temp.
Instruction Manual	1	1	-

Integrity of kit components is guaranteed for up to one year from date of purchase. Reagents are routinely tested on a lot-to-lot basis to ensure they provide the highest performance and reliability.

[1] Ethanol must be added prior to use as indicated on **DNA Wash Buffer** label.

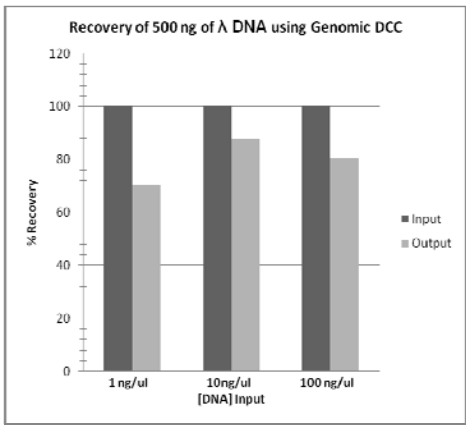
Specifications

- **DNA Purity –** High-quality ($A_{(260/280)}$ ≥ 1.8) high molecular weight DNA ideal for ligation, sequencing, labeling, PCR, microarray, transfection, transformation, and restriction digestion procedures.
- **DNA Size Limits** – Capable of purifying small DNA fragments >50 bp and large sized DNAs >200 kb.
- **DNA Recovery** – Typically, up to 10 µg total DNA per column can be eluted into as ≥10 µl of low salt **DNA Elution Buffer** or water. Recovery of DNA ranges from 70-95%.
- **Sample Sources** – DNA from impure preparations of genomic DNA (e.g., Proteinase K digestions), plasmid DNA (including BAC), viral DNA, and *whole genome amplified* (wga) DNA. Can also be used for the purification of low molecular weight DNA (50 bp to 10 kb) from PCR, endonuclease digestion, post-RT cDNA synthesis, *etc.*.
- **Product Detergent Tolerance** – ≤ 5% Triton X-100, ≤ 5% Tween-20, ≤ 5% Sarkosyl, ≤ 1% SDS.

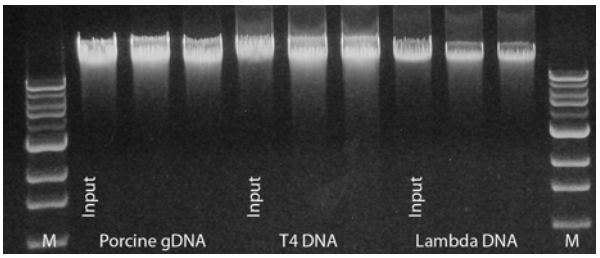
Product Description

The **Genomic DNA Clean & Concentrator™-10 (DCC™)** is for the quick (5 minute) recovery of ultra-pure, large-sized DNA (e.g., genomic, mitochondrial, plasmid (BAC/PAC), viral, phage, (wga)DNA, etc.) from any enzymatic reaction or impure preparation (e.g., Proteinase K digestion). There is no need for organic denaturants, chloroform, or messy precipitations: simply add the specially formulated **ChIP DNA Binding Buffer** to a sample and then transfer the mixture to the supplied **Zymo-Spin™ Column**. Eluted DNA is suitable for sequencing, PCR, endonuclease digestion, and other enzymatic procedures. The product is also compatible with smaller DNAs (50 bp to 10 kb) from PCR, digestions, crude plasmid preparations, cDNA synthesis, etc.

Five minute **Genomic DCC™-10** procedure.



Lambda phage DNA (48.5 kb) is effectively recovered from various concentrations of starting material using the **Genomic DCC™**.



High molecular weight DNA is efficiently purified using the Genomic DCC™-10. Porcine gDNA (~35-50 kb), T4 phage DNA (170 kb), and lambda phage DNA (48.5 kb) were purified (in duplicate) from input material using the Genomic DCC™. Eluted DNAs were analyzed in a 0.8% (w/v) TAE/agarose/EtBr gel (shown above). The size marker "M" is a 1 kb ladder (Zymo Research).

Available Formats

	DCC™ -5	DCC™-25	DCC™-100	DCC™-500	ZR-96 DCC™-5	Genomic DCC™-10	Genomic DCC™-25	ZR-96 Genomic DCC™-5
								
Name	Zymo-Spin™ I & IC	Zymo-Spin™ II & IIC	Zymo-Spin™ V	Zymo-Spin™ VI	Zymo-Spin™ I-96	Zymo-Spin™ IC-XL	Zymo-Spin™ IIC-XL	Zymo-Spin™ I-96-XL
Capacity	5 µg/ prep.	25 µg/ prep.	100 µg/ prep.	500 µg/ prep.	5 µg/ prep.	10 µg/ prep.	25 µg/ prep.	5 µg/ prep.
Elution Vol.	≥ 6 µl	≥ 25 µl	≥ 150 µl	≥ 2 ml	≥ 10 µl	≥ 10 µl	≥ 35 µl	≥ 15 µl
Cat. Nos.	D4003, D4013	D4005, D4033	D4029, D4030	D4031, D4032	D4023, D4024	D4010, D4011	D4064, D4065	D4066, D4067

Typical DCC™ Applications

Post-PCR DNA Clean-up	Efficient desalting of DNA with the removal of DNA polymerases, primers and free dNTPs.
DNA Clean-up From Enzymatic Reactions	Efficient desalting of DNA with the removal of modifying enzymes, RNA polymerases, ligases, kinases, nucleases, phosphatases, endonucleases, *etc.*
Post-Reverse Transcription (RT) & cDNA Clean-up	Efficiently purifies DNA following RT, either as a DNA/RNA complex or as single stranded cDNA following chemical hydrolysis of the RNA template.
Plasmid DNA Clean-up	Efficiently purifies plasmid DNA from "home-made" preparations of cell free lysates or from commercial kits. Plasmid DNA purified and concentrated using the **DCC™** has proven an excellent substrate for high quality DNA sequencing.
Isotope and Dye Removal	Efficiently removes unincorporated fluorescent (*i.e.*, AMCA, FITC, BIO, DIG, Cy3, Cy5, FAM, *etc.*) and radiolabeled dNTP derivatives from DNA following *in vitro* labeling reactions.
Purification of M13 ssDNA	The **DCC™** can be used for the rapid isolation of single stranded M13 phage DNA directly from phage-infected *E. coli* culture supernatant.

- ✓ For purification of short DNA or RNA oligonucleotides ≥16 nt, use the **Oligo Clean & Concentrator (D4060, D4061)**.
- ✓ For ChIP (Chromatin Immunoprecipitation) sample cleanup, use **the ChIP DNA Clean & Concentrator (D5201, D5205)** for high quality DNA from any step in a standard ChIP protocol.
- ✓ For post-cycle sequencing samples, use the **ZR Sequencing DNA Clean-up Kit (D4050, D4051)** for dye blob elimination.
- ✓ For samples containing PCR inhibitors, use the ***OneStep*™ PCR Inhibitor Removal Kit (D6030, D6035)**.

Selected Citations

Marandel, L. et al. (2012). Evolutionary history of *c-myc* in teleosts and characterization of the duplicated *c-myca* genes in goldfish embryos. *Molecular Reproduction and Development*, *79*: 85–96.

Depledge, D.P. et al. (2011). Specific capture and whole-genome sequencing of viruses from clinical samples. PLoS ONE *6*(11)*:* e27805.

Furst, R.W. et al. (2012). Is DNA methylation an epigenetic contribution to transcriptional regulation of the bovine endometrium during the estrous cycle and early pregnancy? *Molecular and Cell Endocrinology, 348* (1), 67-77.

Buffer Preparation

✓ *Before starting:* Add 24 ml 100% ethanol (26 ml 95% ethanol) to the 6 ml **DNA Wash Buffer** concentrate. Add 96 ml 100% ethanol (104 ml 95% ethanol) to the 24 ml **DNA Wash Buffer** concentrate.

Protocol

Note: All centrifugation steps should be performed between 10,000 - 16,000 x g.

1. In a 1.5 ml microcentrifuge tube, add 2-5 volumes of **ChIP DNA Binding Buffer** to each volume of DNA sample[1] (see table below). Mix thoroughly.

Application	DNA Binding Buffer : Sample	Example
Plasmid, genomic DNA (>2 kb)	2 : 1	200 µl : 100 µl
PCR product, DNA fragment	5 : 1	500 µl : 100 µl

2. Transfer mixture to a provided **Zymo-Spin™ IC-XL Column**[2] in a **Collection Tube**.

3. Centrifuge for 30 seconds. Discard the flow-through.

4. Add 200 µl **DNA Wash Buffer** to the column. Centrifuge for 1 minute. Repeat the wash step.

5. Add ≥ 10 µl **DNA Elution Buffer**[3] or water[4] directly to the column matrix and incubate at room temperature for one minute. Transfer the column to a 1.5 ml microcentrifuge tube and centrifuge at for 30 seconds to elute the DNA.

   Ultra-pure DNA is now ready for use.

For **Assistance**, please contact Zymo Research Technical Support at 1-888-882-9682 or e-mail tech@zymoresearch.com.

Notes:

[1] It may be necessary to add RNase A to cell lysates prior to performing the procedure to ensure RNA-free DNA will be recovered in Step 5.

[2] The sample capacity of the column is 1 ml. It may be necessary to load and spin a column multiple times if a sample has a volume larger than 1 ml.

[3] **DNA Elution Buffer**: 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA

[4] Elution of DNA from the column is dependent on pH and temperature. If water is used, make sure the pH is >6.0. Tthe total yield may be improved by eluting the DNA with 60-70 ºC **DNA Elution Buffer**.

Troubleshooting

Low Recovery

- **Improperly Prepared/Stored DNA Wash Buffer**
  Make sure ethanol has been added to the **DNA Wash Buffer** concentrate. Cap the bottle tightly to prevent evaporation over time.
- **Addition of DNA Elution Buffer**
  Add elution buffer directly to the column matrix and not to the walls of the column. Elution buffer requires contact with the matrix for at least 1 minute for large DNA ≥ 10 kb recovery.
- **Incomplete Elution**
  1. DNA elution is dependent on pH, temperature, and time. For large genomic DNA (≥ 50 kb), apply heated elution buffer (60-70 °C) and incubate for several minutes prior to elution.
  2. Sequential elutions may be performed for quantitatively higher recovery but lower final DNA concentration. This is recommended for DNA ≥ 10 kb.

Low A_{260}/A_{230} Ratios

- **Column Tip Contaminated**
  When removing the column from the collection tube, be careful that the tip of the column does not come into contact with the flowthrough. Trace amounts of salt from the flowthrough can contaminate a sample resulting in low A_{260}/A_{230} ratios. Ethanol contamination from the flowthrough can also interfere with DNA elution. Zymo-Spin™ columns are designed for complete elution with no buffer retention or carryover.

Following Clean-up with the DCC™, Multiple Bands Appear in an Agarose Gel

- **Acidification of DNA Loading Dye**
  Most loading dyes do not contain EDTA and will acidify (pH ≤ 4) over time due to some microbial growth. This low pH is enough to cause DNA degradation. Therefore, if water is used to elute the DNA, 6X Loading Dye containing 1 mM EDTA is recommended.

Ordering Information

Product Description	Catalog No.	Kit Size (Preps.)
DNA Clean & Concentrator™-5 (for purification of up to 5 µg DNA per prep.) *Supplied with **uncapped columns***	D4003 D4004	50 200
DNA Clean & Concentrator™-5 (for purification of up to 5 µg DNA per prep.) *Supplied with **capped columns***	D4013 D4014	50 200
ZR-96 DNA Clean & Concentrator™-5 (for 96-well purification of up to 5 µg DNA per well)	D4023 D4024	2 x 96 4 x 96
DNA Clean & Concentrator™-25 (for purification of up to 25 µg DNA per prep.) *Supplied with **uncapped columns***	D4005 D4006	50 200
DNA Clean & Concentrator™-25 (for purification of up to 25 µg DNA per prep.) *Supplied with **capped columns***	D4033 D4034	50 200
DNA Clean & Concentrator™-100 (for purification of up to 100 µg DNA per prep.)	D4029 D4030	25 50
DNA Clean & Concentrator™-500 (for purification of up to 500 µg DNA per prep.)	D4031 D4032	10 20
Oligo Clean & Concentrator™ (for purification of up to 5 µg of oligonucleotides per prep.)	D4060 D4061	50 200
Genomic DNA Clean & Concentrator™-10 (for purification of up to 10 µg genomic DNA per prep.)	D4010 D4011	25 100
Genomic DNA Clean & Concentrator™-25 (for purification of up to 25 µg genomic DNA per prep.)	D4064 D4065	25 100
ZR-96 Genomic DNA Clean & Concentrator™-5 (for 96-well purification of up to 5 µg genomic DNA per well)	D4066 D4067	2 x 96 4 x 96

For Individual Sale	Catalog No.	Size
Chip DNA Binding Buffer	D5201-1-50	50 ml
DNA Wash Buffer (concentrate)	D4003-2-6 D4003-2-24	6 ml 24 ml
DNA Elution Buffer	D3004-4-1 D3004-4-4	1 ml 4 ml
Collection Tubes	C1001-50 C1001-500 C1001-1000	50 tubes 500 tubes 1000 tubes
Zymo-Spin™ IC-XL Columns	C1002-25 C1002-50	25 Columns 50 Columns

DNA PURIFICATION

What is Clean-Spin™ Technology?

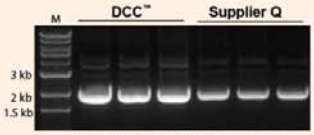
The spin columns from Zymo Research have been designed to ensure complete elution with no binding/wash buffer carryover. The result is ultra-pure inhibitor-free DNA and RNA.

Purify DNA from PCR & other sources

DNA Clean & Concentrator™ (DCC™)

- ✓ Recovery of ultra-pure DNA that is free of salts and contaminants.
- ✓ Small (≥6 µl) elution volume.
- ✓ DNA is ideal for ligation, PCR, Next-Gen sequencing, etc.

Product	Size (Cat. No.)
DNA Clean & Concentrator™-5	50 Preps. (D4013) 200 Preps. (D4014)
ZR-96 DNA Clean & Concentrator™-5	2 x 96 Preps. (D4023) 4 x 96 Preps. (D4024)
Genomic DNA Clean & Concentrator™	25 Preps. (D4010) 100 Preps. (D4011)



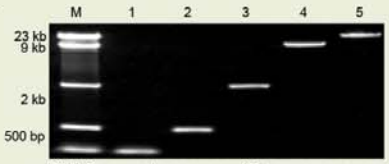
High efficiency DNA recovery with the DCC™-5 compared to Supplier Q .

Boost DNA recoveries from agarose gels to >80%

Zymoclean™ Gel DNA Recovery

- ✓ Rapid (15 min.) recovery of ultra-pure DNA from agarose gels in ≥6 µl.
- ✓ Ultra-pure DNA ideal for DNA ligation, sequencing, etc.
- ✓ Format also available for large DNA >20 kb.

Product	Size (Cat. No.)
Zymoclean™ Gel DNA Recovery Kit	50 Preps. (D4001) 200 Preps. (D4002)
Zymoclean™ Large Fragment DNA Recovery Kit	25 Preps. (D4045) 100 Preps. (D4046)



DNA fragments recovered from an agarose gel using the Zymoclean™ Gel DNA Recovery Kit. Lanes: M: DNA Ladder; 1-5: individual ladder DNA fragments.

Recover transfection-quality plasmid DNA directly from culture

Zyppy™ Plasmid Prep Kits

- ✓ The fastest, simplest method available for purifying high quality plasmid DNA from *E. coli.*
- ✓ Pellet-Free™ procedure omits conventional cell-pelleting and resuspension steps.
- ✓ Transfection quality plasmid DNA directly from culture in under 15 minutes.

Product	Size (Cat. No.)
Zyppy™ Plasmid Miniprep Kit	50 Preps. (D4036) 100 Preps. (D4019) 400 Preps. (D4020) 800 Preps. (D4037)

RNA PURIFICATION

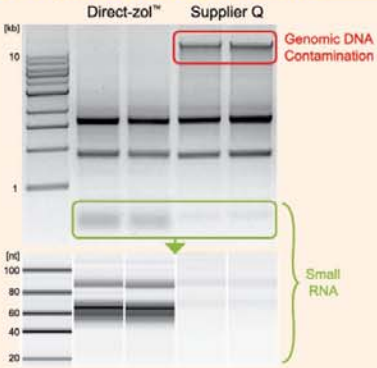
Get RNA *directly* from TRIzol® without phase separation

Direct-zol™ RNA

- ✓ For purification of high-quality small and large RNA *directly* from TRIzol®, TRI Reagent®, or similar.
- ✓ Bypasses phase separation and precipitation procedures allowing for unbiased recovery of miRNA

Product	Size (Cat. No.)
Direct-zol™ RNA MiniPrep	50 Preps. (R2050) 50 Preps. (R2051)* 200 Preps. (R2052) 200 Preps. (R2053)*
96-well and MagBead formats also available!	

DNase I included in all kits.
* Supplied with *TRI-Reagent®*



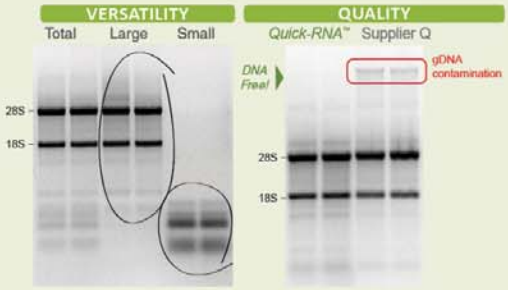
High-quality small and large RNA are effectively recovered with the Direct-zol™ kit. RNA is ***DNA-free***.

Isolate DNA-free RNA from 1 to 10^7 cells in minutes

Quick-RNA™

- ✓ Isolation of total, large, or small RNA – *You decide!*
- ✓ Ultra clean, high-quality RNA from a single cell to 10^7 cells.
- ✓ DNA-free RNA ideal for any downstream application – *DNase I included.*

Product	Size (Cat. No.)
Quick-RNA™ MicroPrep	50 Preps. (R1050) 200 Preps. (R1051)
Quick-RNA™ MiniPrep	50 Preps. (R1054) 200 Preps. (R1055)
ZR-96 *Quick-RNA*™	2 x 96 Preps. (R1052) 4 x 96 Preps. (R1053)



Isolate total, large, or small RNA with the *Quick-RNA*™ kit.

RNA is DNA-free using the *Quick-RNA*™ kit

Purify RNA from enzymatic and labeling reactions in 5 minutes

RNA Clean & Concentrator™

- ✓ Recover ultra-pure RNA in small (≥6 µl) elution volumes.
- ✓ Compatible with TRIzol®, phenol, choloform, and RNase inhibitors (RNAlater®).
- ✓ RNA is ideal for RT-PCR, q-PCR, hybridization, arrays, RNA interference, etc.

Product	Size (Cat. No.)
RNA Clean & Concentrator™-5	50 Preps. (R1015) 200 Preps. (R1016)
RNA Clean & Concentrator™-25	50 Preps. (R1017) 100 Preps. (R1018)
ZR-96 RNA Clean & Concentrator™	2x96 well plates (R1080)
DNA-Free RNA Kit™	50 Preps. (R1013) 200 Preps. (R1014)

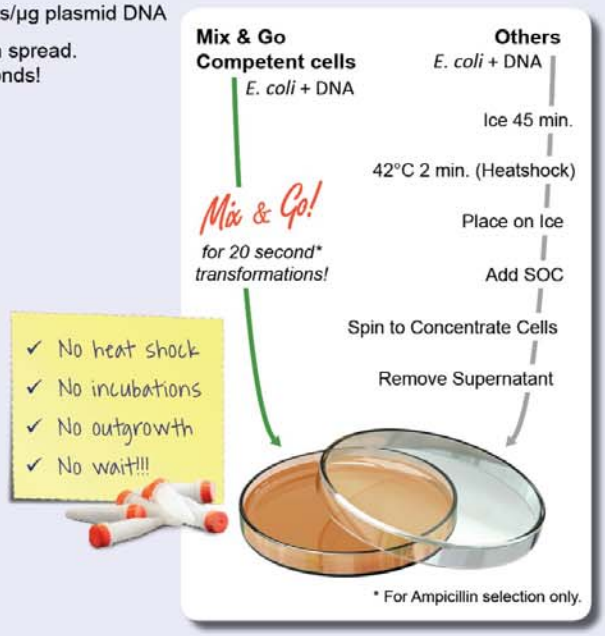
OTHER INNOVATIVE PRODUCTS FROM ZYMO RESEARCH...

Competent cells for transformations *without* heat shock!

Mix & Go! Pre-made Competent *E. Coli*

- ✓ High efficiency: 10^8-10^9 transformants/µg plasmid DNA
- ✓ Just *Mix & Go!* Simply add DNA then spread. Transformation in as little as 20 seconds!

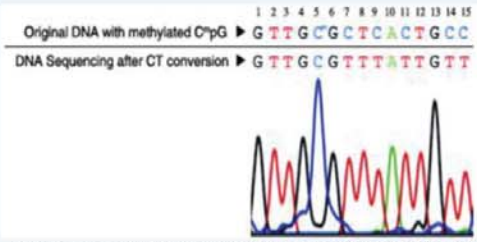
Product	Size (Cat. No.)
Zymo 5α (Same as DH5α)	10 x 100 µl aliquots (T3007) 96 x 50 µl aliquots (T3009) 96 x 50 µl aliquots PCR-plate (T3010)
Zymo 10B (Same as DH10B)	10 x 100 µl aliquots (T3019) 96 x 50 µl aliquots (T3020)
JM109	10 x 100 µl aliquots (T3003) 96 x 50 µl aliquots (T3005)
HB101	10 x 100 µl aliquots (T3011) 96 x 50 µl aliquots (T3013)
C600	10 x 100 µl aliquots (T3015)
TG1	10 x 100 µl aliquots (T3017)



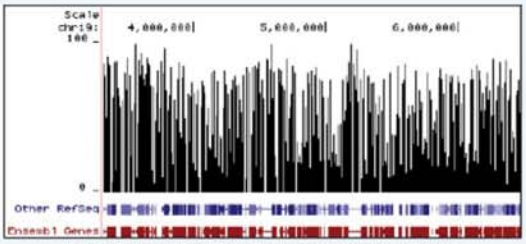
The fastest method for complete bisulfite conversion of DNA

EZ DNA Methylation-Lightning™ Kits

- ✓ The next generation of bisulfite conversion technology by the most cited provider in the industry
- ✓ Guarantees high conversion efficiencies of cytosine (>99.5%)
- ✓ Maintains the highest template integrity following bisulfite conversion
- ✓ Recovered DNA is ideal for PCR, MSP, array, bisulfite, and next-generation sequencing.



DNA Sequencing Results Following Bisulfite Treatment



Methylation Plot From Reduced Representation Bisulfite Sequencing (RRBS)

Product		Size (Cat. No.)
EZ DNA Methylation-Lightning™Kit		50 rxns. (D5030) 200 rxns. (D5031)
EZ-96 DNA Methylation-Lightning™Kit	Shallow-Well Deep-Well	2 x 96 rxns. (D5032) 2 x 96 rxns. (D5033)
EZ-96 DNA Methylation-Lightning™ MagPrep		4 x 96 rxns. (D5046) 8 x 96 rxns. (D5047)