



## Product Manual

Restriction Digest Fragments, Plasmid Cleanup,  
Dilute DNA Concentration, Sequencing Sample Cleanup,  
Genotyping Sample Cleanup

### Omni-Clean™ Gel DNA Purification & Concentration Systems

#### Omni-Clean™ Gel DNA Beads Purification System

Catalog No.: 40-4110-XX

#### Omni-Clean™ Gel DNA Spin Column Purification System

Catalog No.: 40-4120-XX

#### Omni-Clean™ DNA Beads Concentration System

Catalog No.: 40-4130-XX

#### Omni-Clean™ DNA Spin Column Concentration System

Catalog No.: 40-4140-XX



<b>Contents</b>	<b>Page</b>
Omni-Clean™ System	2
<b>Quick Protocols</b>	3
Omni-Clean™ Gel DNA Purification Systems	3
-Gel DNA Beads Purification System	3
-Gel DNA Spin Column Purification System	4
Omni-Clean™ DNA Concentration Systems	5
-DNA Beads Concentration System	5
-DNA Spin Column Concentration System	6
<b>Product Description</b>	7-9
<b>Troubleshooting</b>	10
<b>Omni-Clean™ Gel DNA Purification Systems</b>	11
Materials Supplied	12
Detailed Protocols	13
-Omni-Clean™ Gel DNA Beads Purification System	13
-Omni-Clean™ Gel DNA Spin Column Purification System	14
<b>Omni-Clean™ DNA Concentration Systems</b>	16
Materials Supplied	17
Detailed Protocols	18
-Omni-Clean™ Gel DNA Beads Concentration System	18
-Omni-Clean™ Gel DNA Spin Column Concentration System	19
<b>Appendix</b>	21
<b>Ordering Information</b>	25



## **Omni-Clean™ Gel DNA Purification Systems**

◆ Omni-Clean™ Gel DNA Beads Purification System  
Catalog No: 40-4110-XX

◆ Omni-Clean™ Gel DNA Spin Column Purification System  
Catalog No: 40-4120-XX

◆ Omni-Clean™ DNA Beads Concentration System  
Catalog No: 40-4130-XX

◆ Omni-Clean™ DNA Spin Column Concentration System  
Catalog No: 40-4140-XX

## Omni-Clean™ System

Purification of DNA from agarose gels and concentration of DNA by ethanol precipitation are routine protocols used in all molecular biology laboratories. The Omni-Clean™ system provides optimized reagents for rapid extraction of DNA from agarose gels, and for routine concentration of DNA. The DNA is concentrated, purified and completely salt-free.

### APPLICATION

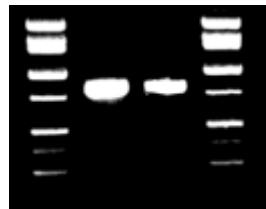
#### Purification of DNA Fragments Excised from Agarose Gels

##### Omni-Clean™ DNA Purification System

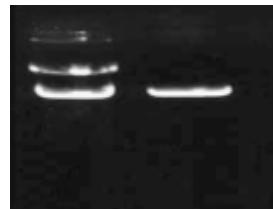
The Omni-Clean™ DNA Purification System takes advantage of the principle that DNA binds to powdered flint glass in the presence of chaotropic salts. This technique provides a rapid and efficient method for the purification of high quality DNA from solutions or agarose gel slices, and is suitable for cloning, sequencing, isotope labeling, and a host of other procedures.

##### Omni-Clean™ System

- Ultra Purified DNA in Less Than 20 minutes
- No Hazardous Reagents
- Suitable for All Molecular Biology Applications
- No More Ethanol Precipitations
- DNA Purification, Concentration and Desalting from Agarose Gel Slices Using Beads or Spin Columns



Lanes 2 and 3 are fragments excised from agarose gel and purified using the Omni-Clean™ column based purification system.



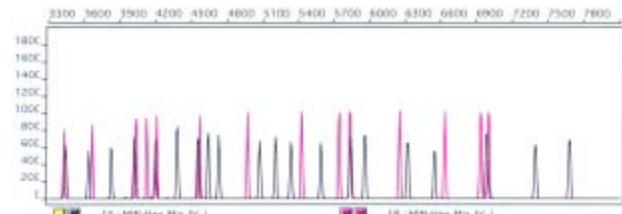
Lane 1 is plasmid extracted using Omni-Pure™ plasmid purification system. Lane 2 is the lower fragment gel purified using the Omni-Clean™ gel glass bead based purification system.

### APPLICATION:

#### Concentration and Cleanup of DNA in Solution

##### Omni-Clean™ DNA Concentration System

The Omni-Clean™ DNA Concentration System yields higher quality DNA than with conventional ethanol precipitation. The samples are completely desalted, purified and concentrated. It is ideal for sequencing and genotyping samples with stringent purity requirements. The electropherograms show samples purified using the Omni-Clean™ system.



Genotyping (top) and sequencing (bottom) electropherogram shows samples purified using the Omni-Clean™ system.

The Omni-Clean™ system is the method of choice to reproducibly yield DNA with stringent purity requirements.



### Ultra Pure Templates

Automated sequencing and genotyping requires high quality template DNA. The Omni-Clean™ system is the method of choice to reproducibly yield ultra high quality DNA

## Omni-Clean™ Gel DNA Beads Purification Systems

### Quick Protocol

#### 20 Minute Ultra Purification of DNA from Agarose Gel Slices

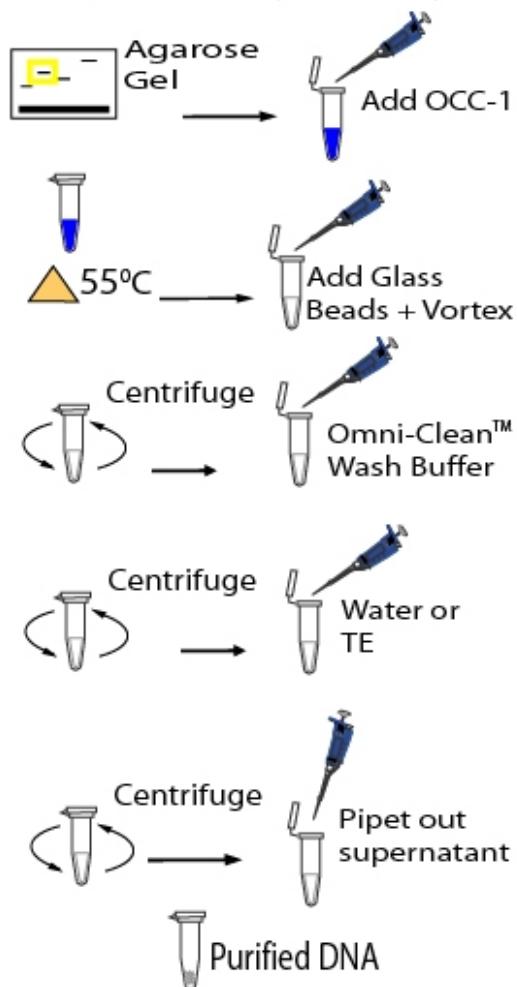
##### Gel DNA Beads Purification System

Catalog No. 40-4110-XX

1. Excise a slice of agarose gel (~3 mm X 5 mm) containing your fragment of interest.
2. Transfer to a 1.5 ml tube and add 300 µl of OCC-1.
3. Incubate at 55°C for 5 minutes. Vortex until gel is completely dissolved.
4. Add 5 µl of glass beads and vortex.
5. Centrifuge at 4K rpm for 30 seconds to pellet glass bead/DNA complex. Discard all traces of supernatant.
6. Re-suspend pellet in 400 µl Omni-Clean™ G3 DNA wash buffer. Centrifuge at 4K rpm for 30 seconds and discard wash buffer.
7. Pipet out any remaining buffer in the tube.
8. Repeat steps 6-7 twice.
9. Add 20 µl water or TE; re-suspend pellet and centrifuge at 6K rpm for 30 seconds.
10. The supernatant contains the purified DNA. Using a pipet collect the supernatant and transfer to a new appropriately labeled tube.

#### Omni-Clean™ Gel DNA Purification System

##### Quick Protocol (Glass Beads)



**i** G3 DNA wash solution is supplied as a 2x concentrate. Dilution with ethanol required prior to use.

**!** OCC-1 contains chaotrope salt blend.

## Omni-Clean™ Gel DNA Spin Column Purification Systems

### Quick Protocol

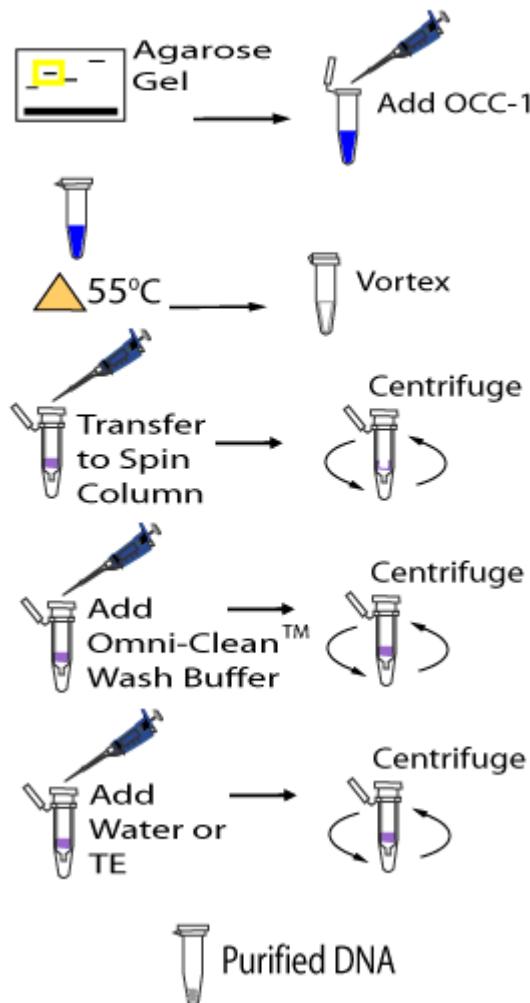
#### 20 Minute Ultra Purification of DNA from Agarose Gel Slices

##### Gel DNA Spin Column Purification System

Catalog No. 40-4120-XX

1. Excise a slice of agarose gel (~3 mm X 5 mm) containing your fragment of interest.
2. Transfer to a 1.5 ml tube and add 300 µl of OCC-1.
3. Incubate at 55°C for 5 minutes. Vortex until gel is completely dissolved.
4. Transfer to a spin column.
5. Centrifuge at 4K rpm for 1 minute. Discard eluent solution.
6. To the spin column add 400 µl Omni-Clean™ G3 DNA wash buffer. Centrifuge at 4K rpm for 1 minute and discard wash buffer.
7. Repeat step 6.
8. Spin empty column at 12K rpm for 1 minute.
9. Replace the collection tube with a new appropriately labeled 1.5 ml tube.
10. Add 50 µl hot water or TE. Allow to remain for 1 minute. Centrifuge at 12K rpm for 1 minute.
11. The eluent collected in the 1.5 ml tube contains the purified DNA.

##### Omni-Clean™ Gel DNA Purification System Quick Protocol (Spin Column)



**!** G3 DNA wash solution is supplied as a 2x concentrate. Dilution with ethanol required prior to use.

**!** OCC-1 contains chaotrope salt blend.

## Omni-Clean™ DNA Beads Concentration Systems

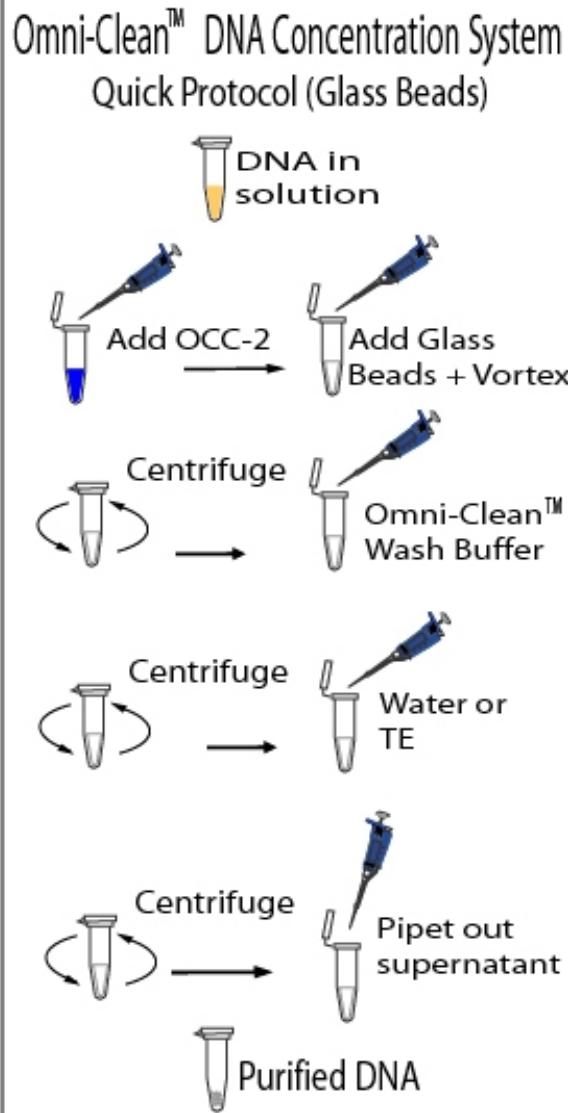
### Quick Protocol

#### 20 Minute Ultra Concentration of DNA

##### DNA Beads Concentration System

Catalog No. 40-4130-XX

1. Determine volume of solution containing DNA that has to be concentrated.
2. Add 2 volumes of OCC-2 and mix.
3. Add 5 µl of glass beads and vortex.
4. Centrifuge at 4K rpm for 30 seconds to pellet glass bead/DNA complex. Discard all traces of supernatant.
5. Re-suspend pellet in 400 µl Omni-Clean™ G3 DNA wash buffer. Centrifuge at 4K rpm for 30 seconds and discard wash buffer.
6. Pipet out any remaining buffer in the tube.
7. Repeat steps 5-6 twice.
8. Add 20 µl water or TE; re-suspend pellet and centrifuge at 6K rpm for 30 seconds.
9. The supernatant contains the purified DNA. Using a pipet collect the supernatant and transfer to a new appropriately labeled tube.



**!** G3 DNA wash solution is supplied as a 2x concentrate. Dilution with ethanol required prior to use.

**!** OCC-2 contains chaotrope salt blend.

## Omni-Clean™ DNA Spin Column Concentration Systems

### Quick Protocol

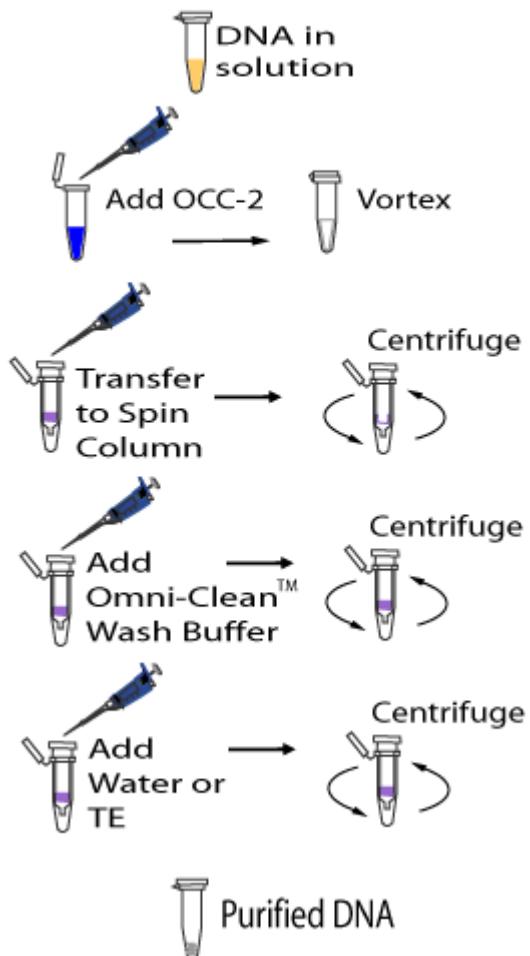
#### 20 Minute Ultra Concentration of DNA

##### DNA Spin Column Concentration System

Catalog No. 40-4140-XX

1. Determine volume of solution containing DNA that has to be concentrated.
2. Add 2 volumes of OCC-2 and mix.
3. Transfer to a spin column.
4. Centrifuge at 4K rpm for 1 minute. Discard eluent solution.
5. To the spin column add 400 µl Omni-Clean™ G3 DNA wash buffer. Centrifuge at 4K rpm for 1 minute and discard wash buffer.
6. Repeat step 5.
7. Spin empty column at 12K rpm for 1 minute.
8. Replace the collection tube with a new appropriately labeled 1.5 ml tube.
9. Add 50 µl hot water or TE. Allow to stand for 1 minute. Centrifuge at 12K rpm for 1 minute.
10. The eluent collected in the 1.5 ml tube contains the purified DNA.

##### Omni-Clean™ DNA Concentration System Quick Protocol (Spin Column)



**i** G3 DNA wash solution is supplied as a 2x concentrate. Dilution with ethanol required prior to use.

**!** OCC-2 contains chaotrope salt blend.

## Product Description

### Introduction

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The Omni-Clean™ DNA purification system is based on the observation that DNA will bind tightly to glass particles in the presence of high concentrations of chaotropic salts (1-4). This purification system is entirely optimized by the formulation of chaotropic salt blends to yield ultra pure DNA from agarose gel in less than 30 minutes and DNA concentration in less than 10 minutes.

With spin column technology the glass is in the form of a thin membrane that is embedded into a small column that can be spun in a centrifuge. In the basic procedure, DNA is applied to the glass beads or column under denaturing conditions in the presence of chaotropic salt. After a brief spin in a centrifuge, the solution passes into a collection tube, leaving the DNA tightly bound to the glass membrane. Following a series of brief washes, the purified DNA can be eluted in a small volume of low salt buffer or sterile water. This procedure is applicable for the purification of DNA from dirty plasmid preps, removal of unwanted products from PCR reactions or enzyme digests and especially for the extraction of DNA fragments from agarose gels. The spin column method is rapid, easy and is ideally suited for handling multiple samples. The purified DNA is ready to use for any application, e.g., PCR, restriction enzyme digests, cloning, sequence analysis, labeling, etc. It should also be noted that an advantage of the spin column method over the glass bead procedure is that there is no possibility of particulate matter carry over during the final elution step. This makes it the method of choice when the purified DNA is to be used for microinjection or capillary electrophoresis.

### DNA Binding to Glass Beads and Glass Membranes

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During the initial binding step, only DNA is bound to the glass membrane. Contaminants that may be present in the solution, such as primers, salts, proteins, unincorporated nucleotides, agarose, dyes, ethidium bromide and detergents (e.g., SDS), do not bind to the glass membrane, but are efficiently removed during the washing steps. Any residual buffers that may impede the quantitative elution of the DNA as well as interfere with subsequent enzymatic reactions are removed by an additional high-speed centrifugation step.

### DNA Elution

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In contrast to DNA absorption, DNA elution is enhanced by low salt concentrations and mild alkaline conditions. DNA can be eluted with Tris Buffer or water. Elution is most efficient between pH 7.0 and 8.5. When using water to elute, make sure that the pH is within this range. Water that has been standing for a long period of time may have pH values as low as 5.5-6 due to absorption of CO<sub>2</sub>. Usually, boiling the water 2-3 minutes in a microwave will remove dissolved CO<sub>2</sub> and brings it within an acceptable pH range. It should be noted, however, that DNA stored in pure water is more sensitive to degradation than when stored in buffered solutions. Therefore, it is recommended that DNA eluted with water be stored at -20°C. Elution with buffers containing EDTA should be avoided because EDTA can interfere with subsequent enzyme reactions.

For efficient elution with 50 µl, the elution buffer or water should be warmed to 60°C and allowed to remain on the column for 1 minute prior to centrifugation. However, elution can also be done with as little as 30 µl of elution buffer or water. When using less than 50 µl it is important to apply the elution buffer directly to the center of the glass membrane and allow it to sit for at least 2 minutes. To ensure quantitative elution it is recommended that following centrifugation the eluent should be reapplied to the column, allowed to sit for 1 minute, and centrifuged a second time.

## Gel Electrophoresis of PCR Product

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Gel electrophoresis of PCR products is the standard method for analyzing reaction quality and yield. PCR products can range up to 10 kb in length, but the majority of amplifications are at 1 kb and below. Agarose electrophoresis is the classical method to analyze amplification products from 150 bp to greater than 10 kb. Polyacrylamide gel electrophoresis should be used for resolution of short fragments in the range of 100 bp to 500 bp when discrimination of a 10 bp difference is required.

PAGE gels for PCR products can be formulated with the amount of cross-linker chosen to give pore sizes optimal for the size of DNA fragment desired. Gels are most often stained in ethidium bromide, even though the fluorescence of this stain is quenched by polyacrylamide, which decreases sensitivity 2-5 fold. This decrease in sensitivity generally does not present a problem, because most PCR reactions yield product levels in the microgram range and ethidium bromide will detect as little as 1/10 of this amount. Polyacrylamide gels can be stained by silver staining for more sensitive detection.

## Purification of PCR Product

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Various purification methods are available for the purification of PCR products. The selection of one particular method over another is based on the downstream application and the initial robustness of the amplification. Usually no further purification is required for most cloning experiments if a single fragment is amplified, whereas for sequencing applications the amplified product should be purified from the primers and any other minor amplification products.

The preferred method of purification of an amplified fragment is the excision of the fragment band after agarose gel electrophoresis. This method yields the purification of a single fragment; as such care should be taken to excise a gel piece containing a single electrophoretically resolved fragment. The Omni-Clean™ Purification System available from Gene Link can be used for this purpose: Catalog No. 40-4110-XX for the glass bead system, 40-4120-XX for the spin column system and 40-4130-XX for the DNA concentration system.

## Spectrophotometric Determination of DNA Concentration

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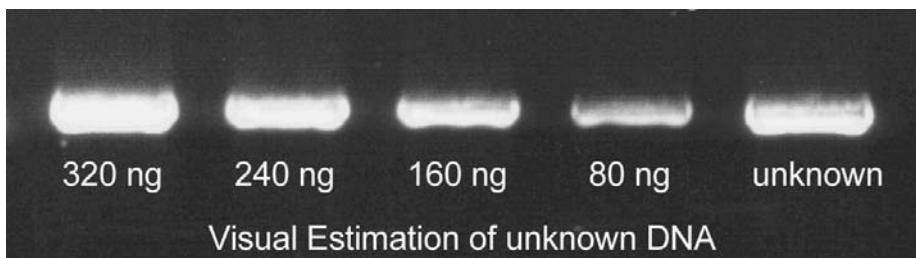
Measuring the optical density (OD) or absorbance at 260 nm ( $A_{260}$ ) in a UV spectrophotometer is a relatively accurate method for calculating the concentration of DNA in an aqueous solution if a standard curve is meticulously prepared. An  $A_{260}$  of 1.0, using a 1 cm path length, corresponds to a DNA concentration of 50 µg/ml for double stranded DNA, 40 µg/ml for single stranded DNA and RNA and 33 µg/ml for oligonucleotides. However, this method is not suitable for determining concentrations of dilute solutions of DNA, as the sensitivity of this method is not very high. For reliable readings, the concentration of double stranded DNA must be greater than 1 µg/ml.

A simple, inexpensive method for the estimation of nanogram quantities of DNA is described in the following section. We recommend the use of agarose gel electrophoresis for routine approximate determination of DNA concentration.

## Estimation of DNA Concentration by Agarose Gel Electrophoresis

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The amount of DNA in a sample may be estimated by running the sample along side of standards containing known amounts of the same-sized DNA fragment. In the presence of ethidium bromide staining, the amount of sample DNA can be visually estimated by comparing the band intensity with that of the known standards.



An unknown amount of a 4 kb DNA fragment was run alongside known quantities (indicated in nanograms) of the same DNA fragment. As estimated by visual comparison with the known standards, the unknown sample contained 240-320 ng of DNA.



Ethidium bromide is a carcinogen. Follow Health and Safety Procedures established by your institution. Follow proper Hazardous Material Disposal procedures established by your institution.

- i • Use 0.1 µg of ethidium bromide solution for each ml of gel volume.

## Agarose Gel Electrophoresis of DNA

Agarose gels are typically run at 20 to 150V. The upper voltage limit is the amount of heat produced. At room temperature about 5 Watts is correct for a minigel (Volts x Amps = Watts). At low voltages migration is linearly proportional to voltage, but long DNA molecules migrate relatively faster in stronger fields. Migration is inversely proportional to the log of the fragment length; a log function also governs migration rate and gel concentration (0.5 to 2% for most purposes). Furthermore, supercoiled/circular DNA molecules migrate at different rates from linear molecules; single-stranded DNA and RNA migrate at similar rates, but usually faster than double-stranded DNA of the same length. Salt in the samples increases conductivity and, hence, migration rate.

The buffers used for most neutral agarose gels (the gel itself and the solution in which it lies) are 1x TAE or 1x TBE. Agarose powder is added to the buffer at room temperature, heated in a microwave and boiled slowly until the powder has dissolved. The gel is cast on a horizontal surface once the agarose has been cooled to ca. 60°C (just cool enough to hold) and 0.1 µg of ethidium bromide solution for each ml of gel volume is added. At times, during removal of the comb, it is possible to tear the bottom of the sample wells, which results in sample leakage upon loading. This can be avoided by removing the comb after the gel has been placed in the running buffer.

- i • Use TAE buffer for most molecular biology agarose gel electrophoresis.

### Recipe

1x TAE Buffer
Agarose Gel Electrophoresis Buffer
40 mM Tris-Acetate pH 7.8
1 mM EDTA

1x TBE
Agarose and Polyacrylamide Gel Electrophoresis Buffer
0.089 M Tris
0.089 M Boric Acid
0.002 M EDTA

## Troubleshooting Guide

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The suggestions in this section should be helpful in resolving the most common problems that may arise during DNA purification. In addition, the members of our Technical Support Services at Gene Link are always happy to answer any questions you may have regarding the procedures in this manual, as well as other applications pertaining to molecular biology.

Problem	Suggestions
Low or no recovery of DNA	<p>Be sure that ethanol was added to the wash buffer concentrate. If not, then repeat the entire procedure with correctly prepared wash buffer.</p> <p>Be sure the pH of the water or buffer used for elution is between 7 and 8.5.</p> <p>Repeated use of electrophoresis buffer can result in elevated pH values. If this is the case, replace with fresh buffer.</p> <p>DNA will remain trapped in un-dissolved gel fragments. Make certain by visual examination that all of the gel has dissolved before adding glass beads or applying the solution to the spin column. The limit of the gel that can be handled by the spin column procedure is 400 mg. For gel slices larger than 400 mg, it is recommended that multiple spin columns be used.</p>
Eluted DNA contains ssDNA which appears as a smear on an analytical gel	The ssDNA can be re-annealed by heating the solution to 95°C for 2 minutes and allowing the tube to cool slowly to room temperature.
There appears to be ethanol in the eluted DNA sample	Following the final wash step, be sure to spin the column at maximum speed for 1 minute to remove the last traces of ethanol.

## References

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1. Vogelstein, B., and Gillespie, D. (1979) Preparative and analytical purification of DNA from agarose. Proc. Natl. Acad. Sci. USA 76, 615-619.
2. Cashion, P., Sathe, G., Javed, A. and Kuster, J. (1980) Hydrophobic affinity chromatography of nucleic acids and proteins. Nucl. Acids Res. 8, 1167-1185.
3. Cashion, P., Javed, A., Harrison, D., Seeley, J., Lentini, V. and Sathe, G. (1981) Hydrophobic chromatography of nucleic acids and proteins on tritylated agarose. In Gene amplification and analysis, Vol. 2. Analysis of nucleic acids by enzymatic methods. Eds. Chirikjian, J.G. and Papas, T.S. Elsevier North Holland, pp. 551-563.
4. Melzak, K.A. et al. (1996) Template preparation and post-PCR clean up. J. Colloid Interface Sci. (USA) 181, 635-644.
5. Sambrook, J. et al., Eds. (1989) Molecular cloning: a laboratory manual, 2nd ed., Cold Spring Harbor Laboratory Press



## Omni-Clean™ Gel DNA Purification Systems

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♦ Omni-Clean™ Gel DNA Beads Purification System  
Catalog No: 40-4110-XX

♦ Omni-Clean™ Gel DNA Spin Column Purification System  
Catalog No: 40-4120-XX

## Materials Supplied

### Omni-Clean™ Gel DNA Purification

	Product	Catalog No.	Size*
<input type="checkbox"/>	Omni-Clean™ Gel DNA Beads Purification System	40-4110-10	100
<input type="checkbox"/>	Omni-Clean™ Gel DNA Beads Purification System	40-4110-50	500
<input type="checkbox"/>	Omni-Clean™ Gel DNA Spin Column Purification System	40-4120-10	100
<input type="checkbox"/>	Omni-Clean™ Gel DNA Spin Column Purification System	40-4120-50	500

\*Unit of size is purifications performed.

### Omni-Clean™ Gel DNA Beads Purification System

Product	Catalog No.	Size	Catalog No.	Size
	<input type="checkbox"/>		<input type="checkbox"/>	
Omni-Clean™ Gel DNA Beads Purification System	40-4110-10	100	40-4110-50	500
<b>Materials Supplied</b>				
OCC-1 (Chaotrope Blend 1)	40-4111-30	30 ml	40-4111-75	2 x 75 ml
Glass Beads	40-4112-75	0.75 ml	40-4112-15	2 x 1.5 ml
G3-DNA Wash buffer; 2x concentrate*	40-4026-06	60 ml*	40-4026-15	2 x 150 ml

### Omni-Clean™ Gel DNA Spin Column Purification System

Product	Catalog No.	Size	Catalog No.	Size
	<input type="checkbox"/>		<input type="checkbox"/>	
Omni-Clean™ Gel DNA Spin Column Purification System	40-4120-10	100	40-4120-50	500
<b>Materials Supplied</b>				
OCC-1 (Chaotrope Blend 1)	40-4111-30	30 ml	40-4111-75	2 x 75 ml
Spin Columns	40-4121-01	100	40-4121-05	500
G3-DNA Wash buffer; 2x concentrate*	40-4026-06	60 ml*	40-4026-15	2 x 150 ml

### \*G3-DNA Wash Solution 2x Concentrate Dilution

#### Dilution Required Prior To Use

Product	Catalog No.	Size	Volume of Absolute Ethanol to Add
*G3-DNA Wash solution; 2x concentrate supplied	40-4026-06	60ml	60 ml
*G3-DNA Wash solution; 2x concentrate supplied	40-4026-15	150 ml	150 ml

## Detailed Protocol

### 20 Minute Gel DNA Ultra Purification Protocol

#### Purification of DNA from Agarose Gels

1. Run DNA on an agarose gel in TAE or TBE buffer.
2. Visualize the DNA bands using a hand-held long-wavelength UV illuminator.
3. Cut the band of interest out of the gel, carrying over a minimal amount of excess agarose.
4. Weigh the gel slice in a 1.5 ml tube. Add 3 volumes of OCC-1 solution per gram of gel. For example, if the gel slice weighs 100 mg, then add 300 µl of OCC-1 solution.
5. Incubate at 55°C, mixing frequently until the agarose is totally dissolved.
6. Mix the glass bead solution well by vortexing and add 5 µl of the glass bead solution to the dissolved agarose/DNA solution. This amount of glass beads is sufficient to bind 5 µg of DNA. If more DNA is present, then add 1 µl of glass bead solution for each additional µg of DNA.
7. Mix the solution by vortexing and keep on ice for 5 minutes, mixing occasionally. Centrifuge at 4K rpm for 30 seconds and discard the supernatant.
8. Resuspend the glass bead pellet in 400 µl of Omni-Clean™ G3 DNA wash solution using a pipette. (Due to the presence of ethanol, vortexing the glass bead pellet until it is completely in suspension may take a while.) Centrifuge for 30 seconds and wash the pellet 2 more times with the same volume of Omni-Clean™ G3 DNA wash solution. After removing the final wash solution, centrifuge the tube one more time and remove the residual alcohol with a pipette. For efficient elution of the DNA, it is important to remove the last traces of alcohol by leaving the tube open to the air for 10 minutes.
9. Resuspend the pellet in at least 10-20 µl of H<sub>2</sub>O or low salt buffer by vortexing. Centrifuge for 30 seconds at 6K rpm and collect the eluted DNA in the supernatant. Repeat this step one more time to ensure efficient elution of the DNA. For more efficient elution use hot (~55°C) sterile water or TE.
10. The DNA is now ready for ligation, restriction enzyme digestion, radiolabeling, etc.

**i** • Use filter barrier pipette tips to prevent cross contamination.

**i** • Dilute G3 concentrate DNA wash solution prior to use.

#### Recipe

G3 DNA Wash Concentrate	
Dilution of G3 Concentrate With 100 % Ethanol Required Prior to Use	
G3 2x Concentrate	100% Ethanol To Add
60 ml	60 ml
150 ml	150 ml

#### Key Steps

- Ensure complete dissolution of agarose gel slice.
- It is essential to remove all traces of G3 DNA wash solution prior to elution of DNA.



- Solution G3 after reconstitution contains ethanol and is flammable.

## Detailed Protocol

### 20 Minute Gel DNA Ultra Purification Protocol

#### Purification of DNA from Agarose Gels

1. Run DNA on an agarose gel in TAE or TBE buffer.
2. Visualize the DNA bands using a hand-held long-wavelength UV illuminator.
3. Cut the band of interest out of the gel, carrying over a minimal amount of excess agarose.
4. Weigh the gel slice in a 1.5 ml tube. Add 3 volumes of OCC-1 solution per gram of gel. For example, if the gel slice weighs 100 mg, then add 300 µl of OCC-1 solution.
5. Incubate at 55°C, mixing frequently until the agarose is totally dissolved.
6. Transfer the dissolved agarose gel solution to a spin column properly placed with a waste collection tube. One spin column binds 10 µg of DNA. If more DNA is present, then consider applying the solution to 2 spin columns.
7. Centrifuge at 4K rpm for 1 minute and discard the eluent. DNA is bound to the column membrane.
8. Add 400 µl of Omni-Clean™ G3 DNA wash solution to the spin column. Centrifuge at 4K rpm for 1 minute, discarding the eluent. Repeat 1 more times with the same volume of Omni-Clean™ G3 DNA wash solution. After removing the final wash solution, centrifuge the spin column one more time at 12K rpm 1 minute without any solution added. This 'empty' spin removes the residual alcohol. For efficient elution of the DNA, it is important to remove the last traces of alcohol.
9. For elution of DNA, place the spin column in a clean 1.5 ml tube and directly add 50 µl of H<sub>2</sub>O or TE to the spin column membrane. Allow the eluent to remain for 1 minute. Centrifuge for 1 minute at 12K rpm. The DNA is eluted into the 1.5 ml tube. Repeat this step one more time to ensure efficient elution of the DNA. For more efficient elution use hot (~55°C) sterile water or TE.
10. The DNA is now ready for ligation, restriction enzyme digestion, radiolabeling, etc.

**i** • Use filter barrier pipette tips to prevent cross contamination.

**i** • Dilute G3 concentrate DNA wash solution prior to use.

#### ● Recipe

G3 DNA Wash Concentrate	
Dilution of G3 Concentrate With 100 % Ethanol Required Prior to Use	
G3 2x Concentrate	100% Ethanol To Add
60 ml	60 ml
150 ml	150 ml

#### ● Key Steps

- Ensure complete dissolution of agarose gel slice.
- It is essential to remove all traces of G3 DNA wash solution prior to elution of DNA.



- Solution G3 after reconstitution contains ethanol and is flammable.

### **Additional notes on DNA Purification with glass beads:**

If DNA is being isolated from relatively large gel slices, then an additional wash with OCC-1 solution alone should be performed prior to rinsing with the Omni wash buffer. This step will remove any residual agarose. The amount of OCC-1 solution to use in the additional step should be about 10 times the volume of the glass bead pellet.

The OCC-1 solution may turn slightly yellow with age, which does not affect its performance. Very old solutions should be checked for changes in pH. A pH of 8.5 or above inhibits the binding of DNA to glass.

A 10% sodium sulfite solution can be used to remove the yellow staining that results from spills of the OCC-1 solution.

### **Additional notes on DNA purification with spin columns:**

1. During the initial binding step, only DNA is absorbed to the glass membrane. Contaminants that may be present in the solution, such as primers, salts, proteins, unincorporated nucleotides, agarose, dyes, ethidium bromide, and detergents (e.g., SDS) do not bind to the glass membrane, but are efficiently removed during the washing steps with buffers. Any residual buffer G3, which may impede the quantitative elution of the DNA as well as interfere with subsequent enzymatic reactions, is removed by an additional high-speed centrifugation step.
2. In contrast to DNA absorption, DNA elution is enhanced by low salt concentrations and mild alkaline conditions. DNA can be eluted with low salt buffer (10 mM Tris-Cl, pH 8.0) or water. Elution is most efficient between pH 7.0 and 8.5. When using water to elute, make sure that the pH is within this range. Water that has been standing for a long period of time may have pH values as low as 5.5-6 due to absorption of CO<sub>2</sub>. Usually, boiling the water 2-3 minutes in a microwave will remove dissolved CO<sub>2</sub> and brings it within an acceptable pH range. It should be noted, however, that DNA stored in pure water is more sensitive to degradation than when stored in buffered solutions. Therefore, it is recommended that DNA eluted with water be stored at -20°C. Elution with buffers containing EDTA should be avoided because EDTA can interfere with subsequent enzyme reactions.
3. For efficient elution with 50 µl, the elution buffer or water should be warmed to 60°C and allowed to remain on the column for 1 minute prior to centrifugation. However, elution can also be done with as little as 30 µl of elution buffer or water. When using less than 50 µl it is important to apply the elution buffer directly to the center of the glass membrane and allow it to sit for at least 2 minutes. To ensure quantitative elution it is recommended that following centrifugation, the eluent should be reapplied to the column, allowed to sit for 1 minute, and centrifuged a second time.



## Omni-Clean™ DNA Concentration Systems

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◆ Omni-Clean™ DNA Beads Concentration System  
Catalog No: 40-4130-XX

◆ Omni-Clean™ DNA Spin Column Concentration System  
Catalog No: 40-4140-XX

## Materials Supplied

### Omni-Clean™ DNA Purification and Concentration Systems

	Product	Catalog No.	Size*
<input type="checkbox"/>	Omni-Clean™ DNA Beads Concentration System	40-4130-10	100
<input type="checkbox"/>	Omni-Clean™ DNA Beads Concentration System	40-4130-50	500
<input type="checkbox"/>	Omni-Clean™ DNA Spin Column Concentration System	40-4140-10	100
<input type="checkbox"/>	Omni-Clean™ DNA Spin Column Concentration System	40-4140-50	500

\*Unit of size is purifications performed.

### Omni-Clean™ DNA Beads Concentration System

Product	Catalog No.	Size	Catalog No.	Size
	<input type="checkbox"/>		<input type="checkbox"/>	
Omni-Clean™ DNA Beads Concentration System	40-4130-10	100	40-4130-50	500
<b>Materials Supplied</b>				
OCC-2 (Chaotrope Blend 2)	40-4131-30	30 ml	40-4131-75	2 x 75 ml
Glass Beads	40-4112-75	0.75 ml	40-4112-15	2 x 1.5 ml
G3 DNA Wash buffer; 2x concentrate*	40-4026-06	60 ml*	40-4026-15	2 x 150 ml*

### Omni-Clean™ DNA Spin Column Concentration System

Product	Catalog No.	Size	Catalog No.	Size
	<input type="checkbox"/>		<input type="checkbox"/>	
Omni-Clean™ DNA Spin Column Concentration System	40-4140-10	100	40-4140-50	500
<b>Materials Supplied</b>				
OCC-2 (Chaotrope Blend 2)	40-4131-30	30 ml	40-4131-75	2 x 75 ml
Spin Columns	40-4121-01	100	40-4121-05	500
G3 DNA Wash buffer; 2x concentrate*	40-4026-06	60 ml*	40-4026-15	2 x 150 ml*

### \*G3- DNA Wash Solution 2x Concentrate Dilution

#### Dilution Required Prior To Use

Product	Catalog No.	Size	Volume of Absolute Ethanol To Add
*G3-DNA Wash solution; 2x concentrate supplied	40-4026-06	60 ml	60 ml
*G3-DNA Wash solution; 2x concentrate supplied	40-4026-15	150 ml	150 ml

## Detailed Protocol

### 20 Minute DNA Concentration & Purification Protocol

#### Concentration of DNA using Glass Beads

1. Determine volume of solution containing DNA that has to be concentrated.
2. Add 2 volumes of OCC-2 and mix.
3. Mix the glass bead solution well by vortexing and add 5 µl of the glass bead solution to the dissolved DNA. This amount of glass beads is sufficient to bind 5 µg of DNA. If more DNA is present, then add 1 µl of glass bead solution for each additional µg of DNA.
4. Mix the solution by vortexing and keep on ice for 5 minutes, mixing occasionally. Centrifuge at 4K rpm for 30 seconds and discard the supernatant.
5. Resuspend the glass bead pellet in 400 µl of Omni-Clean™ G3 DNA wash solution using a pipette. (Due to the presence of ethanol, vortexing the glass bead pellet until it is completely in suspension may take a while.) Centrifuge at 4K rpm for 30 seconds and wash the pellet 2 more times with the same volume of Omni-Clean™ G3 DNA wash solution. After removing the final wash solution, centrifuge the tube one more time and remove the residual alcohol with a pipette. For efficient elution of the DNA, it is important to remove the last traces of alcohol by leaving the tubes open to the air for 10 minutes.
6. Resuspend the pellet in at least 10-20 µl of H<sub>2</sub>O or low salt buffer by vortexing. Centrifuge for 30 seconds at 6K rpm and collect the eluted DNA in the supernatant. Repeat this step one more time to ensure efficient elution of the DNA. For more efficient elution use hot (~55°C) sterile water or TE.
7. The DNA is now ready for ligation, restriction enzyme digestion, radiolabeling, etc.



• Use filter barrier pipette tips to prevent cross contamination.



• Dilute G3 concentrate DNA wash solution prior to use.

#### Recipe

G3 DNA Wash Concentrate	
Dilution of G3 Concentrate With 100 % Ethanol Required Prior to Use	
G3 2x Concentrate	100% Ethanol To Add
60 ml	60 ml
150 ml	150 ml



• It is essential to remove all traces of G3 DNA wash solution prior to elution of DNA.



• Solution OCC-2 contains guanidine hydrochloride, which can form highly reactive compounds when combined with bleach (sodium hypochlorite). If liquid containing this reagent is spilled, clean with water and, if necessary, an appropriate laboratory detergent.

• Solution G3 after reconstitution contains ethanol and is flammable.

## Detailed Protocol

### 20 Minute Spin Column DNA Concentration Protocol

#### Concentration of DNA using Spin Column

1. Determine volume of solution containing DNA that has to be concentrated.
2. Add 2 volumes of OCC-2 and mix.
3. Transfer the solution to a spin column properly placed with a waste collection tube. One spin column binds 10 µg of DNA. If more DNA is present, then consider applying the solution to 2 spin columns.
4. Centrifuge at 4K rpm for 1 minute and discard the eluent. DNA is bound to the column membrane.
5. Add 400 µl of Omni-Clean™ G3 DNA wash solution to the spin column. Centrifuge at 4K rpm for 1 minute, discarding the eluent. Repeat 1 more times with the same volume of Omni-Clean™ G3 DNA wash solution. After removing the final wash solution, centrifuge the spin column one more time at 12K rpm for 1 minute without any solution added. This 'empty' spin removes the residual alcohol. For efficient elution of the DNA, it is important to remove the last traces of alcohol.
6. For elution of DNA, place the spin column in a clean 1.5 ml tube and directly add 50 µl of H<sub>2</sub>O or TE to the spin column membrane. Allow the eluent to remain for 1 minute. Centrifuge for 1 minute at 12K rpm. The DNA is efficiently eluted in the 1.5 ml tube. Repeat this step one more time to ensure efficient elution of the DNA. For more efficient elution use hot (~55°C) sterile water or TE. The DNA is now ready for ligation, restriction enzyme digestion, radiolabeling, etc.

**i** •Use filter barrier pipette tips to prevent cross contamination.

**i** •Dilute G3 concentrate DNA wash solution prior to use.

#### Recipe

G3 DNA Wash Concentrate	
Dilution of G3 Concentrate With 100 % Ethanol Required Prior to Use	
G3 2x Concentrate	100% Ethanol To Add
60 ml	60 ml
150 ml	150 ml

**i** •It is essential to remove all traces of G3 DNA wash solution prior to elution of DNA.

**!** •Solution OCC-2 contains guanidine hydrochloride, which can form highly reactive compounds when combined with bleach (sodium hypochlorite). If liquid containing this reagent is spilled, clean with water and, if necessary, an appropriate laboratory detergent.  
•Solution G3 after reconstitution contains ethanol and is flammable.

### **Additional notes on DNA Purification with glass beads:**

If DNA is being isolated from relatively large gel slices, then an additional wash with OCC-1 solution alone should be performed prior to rinsing with the Omni wash buffer. This step will remove any residual agarose. The amount of OCC-1 solution to use in the additional step should be about 10 times the volume of the glass bead pellet.

The OCC-1 solution may turn slightly yellow with age, which does not affect its performance. Very old OCC-1 solutions should be checked for changes in pH. A pH of 8.5 or above inhibits the binding of DNA to glass.

A 10% sodium sulfite solution can be used to remove the yellow staining that results from spills of the OCC-1 solution.

### **Additional notes on DNA purification with spin columns:**

1. During the initial binding step, only DNA is absorbed to the glass membrane. Contaminants that may be present in the solution, such as primers, salts, proteins, unincorporated nucleotides, agarose, dyes, ethidium bromide, and detergents (e.g., SDS) do not bind to the glass membrane, but are efficiently removed during the buffer washing steps. Any residual buffer G3, which may impede the quantitative elution of the DNA as well as interfere with subsequent enzymatic reactions, is removed by an additional high-speed centrifugation step.
2. In contrast to DNA absorption, DNA elution is enhanced by low salt concentrations and mild alkaline conditions. DNA can be eluted with either low salt buffer (10 mM Tris-Cl, pH 8.0) or water. Elution is most efficient between pH 7.0 and 8.5. When using water to elute, make sure that the pH is within this range. Water that has been standing for a long period of time may have pH values as low as 5.5-6 due to absorption of CO<sub>2</sub>. Usually, boiling the water 2-3 minutes in a microwave will remove dissolved CO<sub>2</sub> and bring it within an acceptable pH range. It should be noted, however, that DNA stored in pure water is more sensitive to degradation than when stored in buffered solutions. Therefore, it is recommended that DNA eluted with water be stored at -20°C. Elution with buffers containing EDTA should be avoided because EDTA can interfere with subsequent enzyme reactions.
3. For efficient elution with 50 µl, the elution buffer or water should be warmed to 60°C and allowed to remain on the column for 1 minute prior to centrifugation. However, elution can also be done with as little as 30 µl of elution buffer or water. When using less than 50 µl it is important to apply the elution buffer directly to the center of the glass membrane and allow it to sit for at least 2 minutes. To ensure quantitative elution it is recommended that following centrifugation, the eluent should be reapplied to the column, allowed to sit for 1 minute, and centrifuged a second time.

## Appendix

### PCR Components and Analysis

PCR buffer conditions vary and it is imperative to optimize buffer conditions for each amplification reaction. At Gene Link most amplification reactions have been optimized to work with the following standard buffer condition, unless otherwise indicated.

### dNTP Concentration

Standard dNTP concentration of 0.2 mM of each base is used. See section on PCR additives when dNTP concentration is changed.

### Recipe

Standard Gene Link PCR Buffer Composition	
10x PCR buffer	1x PCR buffer
100 mM Tris-HCl pH 8.3	10 mM
500 mM KCl	50 mM
15 mM MgCl <sub>2</sub>	1.5 mM
0.01% Gelatin	0.001%

### Recipe

2.0 mM dNTP Stock Solution Preparation*	
Component	Volume
100 mM dGTP	100 µl
100 mM dATP	100 µl
100 mM dTTP	100 µl
100 mM dCTP	100 µl
Water	4.6 ml
Total Volume	5 ml

\*Aliquot and freeze

### MgCl<sub>2</sub> Concentration

The concentration of Mg<sup>2+</sup> will vary from 1-5 mM, depending upon primers and substrate. Since Mg<sup>2+</sup> ions form complexes with dNTPs, primers and DNA templates, the optimal concentration of MgCl<sub>2</sub> has to be selected for each experiment. Low Mg<sup>2+</sup> ion concentration results in a low yield of PCR product, and high concentrations increase the yield of non-specific products and promote mis-incorporation. Lower Mg<sup>2+</sup> concentrations are desirable when fidelity of DNA synthesis is critical. The recommended range of MgCl<sub>2</sub> concentration is 1-4 mM, under the standard reaction conditions specified. At Gene Link, using the standard PCR buffer with KCl, a final dNTP concentration of 0.2 mM, a MgCl<sub>2</sub> concentration of 1.5 mM is used in most cases. If the DNA samples contain EDTA or other chelators, the MgCl<sub>2</sub> concentration in the reaction mixture should be raised proportionally. Given below is a MgCl<sub>2</sub> concentration calculation and addition table using a stock solution of 25 mM MgCl<sub>2</sub>.

MgCl <sub>2</sub> Concentration & Addition Table								
Final concentration of MgCl <sub>2</sub> in 50 µl reaction mix, (mM)	1.0	1.25	1.5	1.75	2.0	2.5	3.0	4.0
Volume of 25 mM MgCl <sub>2</sub> , (µl)	2	2.5	3	3.5	4	5	6	8

### Primer Concentration

The final concentration of primers in a PCR reaction is usually 0.5 to 1 µM (micromolar). This is equivalent to 0.5 to 1 pmol/µl. For a 100 µl reaction, add 50 to 100 pmols. At Gene Link we use 0.5 pmol/µl in the final reaction.



Always use filter barrier pipette tips to prevent cross contamination

### Genemer™ Reconstitution

**Stock Primer Mix:** Dissolve the supplied 10 nmols of lyophilized Genemer™ in 100 µl sterile TE. The 10 nmols of primers when dissolved in 100 µl will give a solution of 100 µM, i.e., 100 pmols/µl.

### Recipe

TE Buffer pH 7.5 Composition	
1x TE Buffer pH 7.5	
10 mM Tris-HCl pH 7.5	
1 mM EDTA	

**Primer Mix:** Prepare a 10 pmols/µl Primer Mix solution by a ten fold dilution of the stock primer mix.

Example: Add 180 µl sterile TE to a new tube, to this tube add 20 µl of primer stock solution. Label this tube as Primer Mix 10 pmols/µl.

### Amplification Thermal Cycling

**Hot Start:** It is essential to have a 'Hot Start' profile for amplification of any fragment from a complex template like human genomic DNA. Taq polymerase has low activity at room temperature and it is essential to minimize any mis-priming in the first cycle of amplification. A typical hot start profile is given below. Various enzyme preparations are available which are activated by heat in the first cycle. A simple hot start protocol is given below that can be used with regular Taq polymerase. See the section on PCR additives for amplification of products from high GC content templates.

Hot Start		
Step	Time & Temperature	Cycles
Initial Denaturation	95 °C for 5 minutes	1
Annealing	60 °C Hold Infinity	Hold
Comments: Add Taq premix while on hold.		



- Program your thermal cycler instrument with an amplification profile prior to beginning the amplification protocol.
- Consult your appropriate instrument manufacturer's manual.

### Recipe

Typical PCR Premix (/50µl)	
Component	Volume
10x PCR Buffer	5 µl
2.0 mM dNTP mix (each)	5 µl
Primer Mix (10 pmol/µl each) or 2.5 µl of 10 pmol/µl of individual primer (final 25 pmol of each primer/50 µl)	2.5 µl
H <sub>2</sub> O	37.5 µl
<b>Total Volume</b>	<b>50 µl</b>

### Recipe

PCR reaction (/50µl)	
Component	Volume
PCR premix	45 µl
100 ng/µl diluted DNA	1 µl
Hot start and then add	
Taq premix	5 µl

### Recipe

Taq Premix (/50µl)	
Component	Volume
PCR Premix	6 µl
Taq polymerase (5 units/µl)	0.25µl
<b>Add 5 µl/50 µl rxn after initial denaturation</b>	
Use 2.5 units of Taq for 100 µl reactions.	
Taq is usually supplied at a concentration of 5 units/µl	

Typical Amplification File			
Step	Temperature	Time	Cycles
Denaturation	94°C	30 sec.	30
Annealing	*	30 sec.	
Elongation	72°C	30 sec.	
Fill in Extension	72°C	7 minutes	1
Hold	4°C	Infinity	Hold

\*Based on the T<sub>m</sub> of the primers. Usually varies from 50°C to 65°C

PCR Premix Preparation (PP)		
Component	1x 50 µl Rxn.	10x 50 µl Rxns.
Sterile Water	32 µl	320 µl
10x PCR Buffer	4.5 µl	45 µl
2.0 mM dNTP	5 µl	50 µl
10 pmol/µl Primer Mix	2.5 µl	25 µl
Taq Enzyme Mix (EM) See below for preparation	5 µl	50 µl
Template DNA (~500 ng)	1-2 µl	Add 1-2 µl DNA to each tube
Total Volume	50 µl	
Keep on ice during set up. After adding template start PCR File		



- The PCR premix preparation protocol is written considering that more than one amplification reaction will be performed at the same time. If only one reaction is planned then there is no need to prepare the Taq Enzyme Mix (EM).

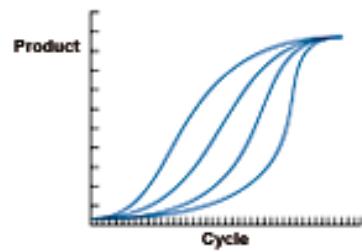
### Recipe

Gene Link PCR Buffer
1x PCR Buffer
10 mM Tris-HCl pH 8.3
50 mM KCl
1.5 mM MgCl <sub>2</sub>
0.001% Gelatin

### **Yield and Kinetics**

The target will be amplified by up to  $10^6$  fold in a successful reaction, but the amplification will usually plateau at 1-10 µg. Thus, 1 pg of target sequence in the reaction is a good place to begin.

PCR reactions produce product in a nonlinear pattern. Amplification follows a typical exponential curve until some saturation point is reached. Generally products will not be further amplified once 1-5 µg has been generated. Saturation by one product of a reaction does not always prevent further amplification of other generally unwanted products. Over-cycling may decrease the quality of an otherwise good reaction. When first optimizing a reaction, it is advisable to take samples every 5 or 10 cycles to determine the number of cycles actually needed.



### **Gel Electrophoresis of PCR Products**

Gel electrophoresis of PCR products is the standard method for analyzing reaction quality and yield. PCR products can range up to 10 kb in length, but the majority of amplifications are at 1 kb and below. Agarose electrophoresis is the classical method to analyze amplification products from 150 bp to greater than 10 kb. Polyacrylamide gel electrophoresis should be used for resolution of short fragments in the range of 100 bp to 500 bp when discrimination of as little as a 10 bp difference is required.

PAGE gels for PCR products can be formulated with the amount of cross-linker chosen to give pore sizes optimal for the size of DNA fragment desired. Gels are most often stained in ethidium bromide, even though the fluorescence of this stain is quenched by polyacrylamide, which decreases sensitivity 2-5 fold. This decrease in sensitivity generally does not present a problem, because most PCR reactions yield product levels in the microgram range, and ethidium bromide will detect as little as 1/10 of this amount. Polyacrylamide gels can be stained by silver staining for more sensitive detection.

### **PCR Additives**

DNA polymerases need to elongate rapidly and accurately to function effectively *in vivo* and *in vitro*, yet certain DNA regions appear to interfere with their progress. One common problem are pause sites, at which DNA polymerase molecules cease elongation for varying lengths of time. Many strong DNA polymerase pauses are at the beginnings of regions of strong secondary structure such as template hairpins (1). Taq polymerase used in PCR suffers the same fate and GC-rich DNA sequences often require laborious work to optimize the amplification assay. The GC-rich sequences possess high thermal and structural stability, presumably because the high duplex melting temperature that permits stable secondary structures to form, thus preventing completion of a faithful replication (2).

Nucleotide analog 7-deaza dGTP is effective in reducing the secondary structure associated with the GC-rich region by reducing the duplex stability (4). Betaine, DMSO and formamide reduce the  $T_m$  and the complex secondary structure, thus the duplex stability (1-5). Tetramethyl ammonium chloride (TMAC) actually increases the specificity of hybridization and increases the  $T_m$ . The use of TMAC is recommended in PCR conditions using degenerate primers.

These PCR additives and enhancing agents have been used to increase the yield, specificity and consistency of PCR reactions. These additives may have beneficial effects on some amplifications and it is impossible to predict which agents will be useful in a particular context and therefore they must be empirically tested for each combination of template and primers.

PCR Additives		
Additive	Purpose & Function	Concentration
7-deaza-2'-deoxyguanosine; 7-deaza dGTP	GC-rich region amplification. Reduces the stability of duplex DNA	Totally replace dGTP with 7-deaza dGTP; or use 7-deaza dGTP: dGTP at 3:1
Betaine (N,N,N-trimethylglycine = [carboxymethyl] trimethyl ammonium)	Reduces $T_m$ facilitating GC-rich region amplification. Reduces duplex stability	Use 3.5 M to 0.1 M Betaine. Be sure to use Betaine or Betaine (mono)hydrate and <b>not</b> Betaine HCl.
BSA (bovine serum albumin)	BSA has proven particularly useful when attempting to amplify ancient DNA or templates which contain PCR inhibitors such as melanin.	BSA concentration of 0.01 $\mu$ g/ $\mu$ l to 0.1 $\mu$ g/ $\mu$ l can be used.
DMSO (dimethyl sulfoxide)	DMSO is thought to reduce secondary structure and is particularly useful for GC-rich templates.	DMSO at 2-10% may be necessary for amplification of some templates, however 10% DMSO can reduce <i>Taq</i> polymerase activity by up to 50% so it should not be used routinely.
Formamide	Reduces secondary structure and is particularly useful for GC-rich templates.	Formamide is generally used at 1-5%. Do not exceed 10%.
Non-ionic detergents, e.g., Triton X-100, Tween 20 or Nonidet P-40 (NP-40)	Non-ionic detergents stabilize <i>Taq</i> polymerase and may also suppress the formation of secondary structure.	0.1-1% Triton X-100, Tween 20 or NP-40 may increase yield but may also increase non-specific amplification. As little as 0.01% SDS contamination of the template DNA (left-over from the extraction procedure) can inhibit PCR by reducing <i>Taq</i> polymerase activity to as low as 10%, however, inclusion of 0.5% Tween-20 or -40 will effectively neutralize this effect.
TMAC (tetramethyl ammonium chloride)	TMAC is used to reduce potential DNA-RNA mismatch and improve the stringency of hybridization reactions. It increases $T_m$ and minimizes mis-pairing.	TMAC is generally used at a final concentration of 15-100 mM to eliminate non-specific priming.

## References

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4. Keith, J. M., Cochran, D.A.E., Lala, G.H., Adams, P., Bryant, D. and Mitchelson, K.R. (2004) Unlocking hidden genomic sequence. Nucl. Acids Res. 32: e35.
5. Owczarzy, R., Dunietz, I., Behlke, M.A., Klotz, I.M. and Walder, J.A. (2003) Thermodynamic treatment of oligonucleotide duplex–simplex equilibria. PNAS, 100:14840-14845.

## Ordering Information

### Omni-Clean™ Gel DNA Purification and Concentration Systems

Product	Catalog No.	Size*
Omni-Clean™ Gel DNA Beads Purification System	40-4110-10	100
Omni-Clean™ Gel DNA Beads Purification System	40-4110-50	500
Omni-Clean™ Gel DNA Spin Column Purification System	40-4120-10	100
Omni-Clean™ Gel DNA Spin Column Purification System	40-4120-50	500
Omni-Clean™ DNA Beads Concentration System	40-4130-10	100
Omni-Clean™ DNA Beads Concentration System	40-4130-50	500
Omni-Clean™ DNA Spin Column Concentration System	40-4140-10	100
Omni-Clean™ DNA Spin Column Concentration System	40-4140-50	500

\*Sample volume for each purification system varies. Each purification yields sufficient quantity for desired applications.

## Related Products Ordering Information

### Omni-Pure™ Plasmid DNA Purification Systems

Product	Catalog No.	Size*
Omni-Pure™ Plasmid DNA Purification System	40-4020-01	100
Omni-Pure™ Plasmid DNA Purification System	40-4020-05	500

\*Sample volume for each purification system varies. Each purification yields sufficient quantity for desired applications.

### Omni-Pure™ DNA & RNA Purification Systems

Product	Catalog No.	Size*
Omni-Pure™ Blood DNA Purification System	40-4010-01	100
Omni-Pure™ Blood DNA Purification System	40-4010-05	500
Omni-Pure™ Blood DNA Purification System	40-4010-10	1000
Omni-Pure™ Tissue DNA Purification System	40-4050-01	100
Omni-Pure™ Tissue DNA Purification System	40-4050-05	500
Omni-Pure™ Tissue DNA Purification System	40-4050-10	1000
Omni-Pure™ Plant DNA Purification System	40-4060-01	100
Omni-Pure™ Plant DNA Purification System	40-4060-05	500
Omni-Pure™ Plant DNA Purification System	40-4060-10	1000
Omni-Pure™ Universal DNA Purification System	40-4070-01	100
Omni-Pure™ Universal DNA Purification System	40-4070-05	500
Omni-Pure™ Universal DNA Purification System	40-4070-10	1000
Omni-Pure™ Viral DNA Purification System	40-3720-01	100
Omni-Pure™ Viral DNA Purification System	40-3720-05	500
Omni-Pure™ Microbial DNA Purification System	40-3700-01	100
Omni-Pure™ Microbial DNA Purification System	40-3700-05	500

\*Sample volume for each purification system varies. Each purification yields sufficient quantity for desired applications.

Visit [www.genelink.com](http://www.genelink.com) for pricing and ordering information.

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