

Wizard® Plus Minipreps DNA Purification System

INSTRUCTIONS FOR USE OF PRODUCTS A7100, A7500, A7510, A7141 AND A7211.

Quick
PROTOCOL

Purification Without a Vacuum Manifold (Using 3ml Luer-Lok® Syringes)

Prepare Cleared Lysate	Culture Volume		
	1–3ml	3–5ml	5–10ml
1. Pellet cells.	1–2 minutes 10,000 × g†	10 minutes 10,000 × g†	10 minutes 1,400 × g
2. Suspend pellet in Cell Resuspension Solution.	200µl	300µl	400µl
3. Add Cell Lysis Solution. Invert 4 times to mix.	200µl	300µl	400µl
4. Add Neutralization Solution. Invert 4 times to mix.*	200µl	300µl	400µl
5. Centrifuge lysate for 5 minutes at 10,000 × g.			

Plasmid DNA Purification

- Remove plunger from 3ml Luer-Lok® syringe (Becton-Dickinson Cat.# 9585). Attach syringe barrel to Luer-Lok® extension of Minicolumn.
- Resuspend resin. Add 1ml resin to each Minicolumn/syringe assembly. Carefully transfer cleared lysate (from Step 5) to resin in each assembly.
- Insert plunger and push resin and lysate into Minicolumn.*

Washing

- Detach syringe from Minicolumn; remove plunger from syringe barrel. Reattach barrel to Minicolumn.
- Add 2ml Column Wash Solution containing ethanol. Insert the plunger and push the Column Wash Solution through the Minicolumn.
- Remove syringe and transfer the Minicolumn to a 1.5ml microcentrifuge tube. Centrifuge at 10,000 × g for 2 minutes.

Elution

- Transfer Minicolumn to a new microcentrifuge tube.
- Add 50µl of Nuclease-Free Water to the Minicolumn and wait 1 minute.
For plasmids ≥10kb, use water preheated to 70°C; for plasmids <20kb, use water preheated to 40°C.
- Centrifuge at 10,000 × g for 20 seconds at room temperature.
- Remove and discard Minicolumn. Store DNA at –20°C or below.

†Maximum speed on a microcentrifuge.

For EndA strains and other modifications, additional protocol information is available in Technical Bulletin #TB117, available from Promega or online at www.promega.com

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Ralph Rapley



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Wood Formation in Trees Nigel J Chaffey, 2002-01-10 Trees are a major component of the biosphere and have played an important part in the world's history and culture. With the modern challenges of global warming and dwindling fossil fuel reserves, trees and in particular their wood can provide solutions. Unfortunately, too little is known about the biology of these plants due largely to a lack of Estrogenic control of hypothalamic GnRH neurons Henryk Urbanski, In female mammals sexual maturation is initiated by a change in the release pattern of pituitary gonadotropins: luteinizing hormone (LH) and follicle stimulating hormone (FSH) and a consequential increase in the secretion of ovarian sex steroids: estradiol and progesterone. Orchestrating these endocrine changes is a diffuse population of hypothalamic neurons that produce the neuropeptide gonadotropin-releasing hormone (GnRH). Although the development and functional integration of GnRH neurons with the rest of the central nervous system is still poorly understood, recent progress has been made on several fronts. For example, there is now evidence that some mammalian species express more than one molecular form of GnRH: GnRH I and GnRH II, and that the two corresponding GnRH neuronal subpopulations may play different roles in the regulation of reproductive function and behavior. Moreover, through the use of transgenic animal models, neuronal fiber tracing, gene expression profiling, and electrophysiological recordings, new insights have been gained into the mechanisms that regulate GnRH release. The focus of this Research Topic is on the positive and negative actions that estrogens exert within the mammalian hypothalamus, especially on the reproductive neuroendocrine axis around the time of the preovulatory LH surge. Our hope is that a deeper understanding of the endocrine interactions between the hypothalamus, pituitary gland, and gonads will provide a solid foundation upon which to develop more effective therapies for pubertal disorders, infertility, and menopause. Exercises for the Molecular Biology Laboratory: Instructor's manual Patrick Guilfoile, 2000

Handbook of Nucleic Acid Purification Dongyou Liu, 2009-01-14 An indispensable roadmap for nucleic acid preparation. Although Friedrich Miescher described the first isolation of nucleic acid in 1869, it was not until 1953 that James Watson and Francis Crick successfully deciphered the structural basis of DNA duplex. Needless to say, in the years since, enormous advances have been made in the study of nucleic acids.

G Protein Signaling Alan V. Smrcka, 2008-02-02 Alan V. Smrcka presents a collection of cutting-edge methods for investigating G protein signaling from a variety of perspectives, ranging from in vitro biochemistry to whole animal studies. Among the readily reproducible techniques presented are those for the purification of G proteins and effectors, enzymes, assays of these purified G proteins and effector enzymes, and for the study of G protein interactions with effectors in intact cells. Additional methods are provided for assaying G protein-coupled receptor structure, function, and localization, and for studying the physiological roles for endogenous G proteins.

G Proteins: Techniques of Analysis David R. Manning, 2020-03-16 Incorporating a bench-top format, *G Proteins: Techniques of Analysis* covers essential methods with a commitment to those techniques of proven and current utility. It offers an in-depth description of protocols

together with theory and representative data It includes expression and functional analysis of G proteins evaluation of covalent modifications and other regulatory phenomena and mapping pathways established among receptors G proteins and effectors Incorporating contributors from key institutions each contributor offers clear instructions to establish a synthesized concise and consistent approach to each chapter which is beneficial to both students and professionals

The Nucleic Acid Protocols Handbook Ralph Rapley, 2000-03-24 A comprehensive treasury of all the key molecular biology methods ranging from DNA extraction to gene localization in situ needed to function effectively in the modern laboratory Each of the 120 highly successful techniques follows the format of the much acclaimed Methods in Molecular Biology™ series providing an introduction to the scientific basis of each technique a complete listing of all the necessary materials and reagents and clear step by step instruction to permit error free execution Included for each technique are notes about pitfalls to avoid troubleshooting tips alternate methods and explanations of the reasons for certain steps all key elements contributing significantly to success or failure in the lab The Nucleic Acid Protocols Handbook constitutes today's most comprehensive collection of all the key classic and cutting edge techniques for the successful isolation analysis and manipulation of nucleic acids by both experienced researchers and those new to the field

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Cloning and Characterization of a Genomic Actin of Naegleria Fowleri Suzanne Marie Gorospe, 1995

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recommendation letters are one of the most vital components of your graduate nursing school application materials as such prospective students should carefully consider who they should select to write a recommendation