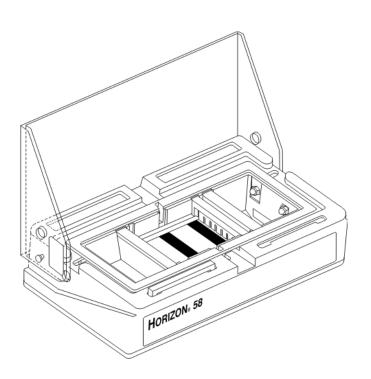


OPERATING MANUAL

Horizon[®] 58 #41060039

Horizontal Electrophoresis Apparatus



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- 4. Sample Loading Buffer
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1.0 BEFORE YOU BEGIN

1.1 IMPORTANT INFORMATION

The **Horizon**[®] **58** (H58) electrophoresis unit is authorized for laboratory research use only. It has not been qualified for use in any human, animal diagnostic or therapeutic application. Use for other than the intended use may be a violation of applicable law.

The H58 is a horizontal format gel electrophoresis apparatus (Figure 1) designed for rapid, high resolution separation of nucleic acids. It is suitable for agarose gel electrophoresis procedures where buffer recirculation is not required.

If the product is used in a manner not specified by Apogee, the protection provided by safety features of the product may be impaired. Please carefully follow the manual's instructions. Do not alter equipment or operate with broken components. Failure to adhere to these directions could result in personal and/or laboratory hazards as well as invalidate the equipment warranty.

1.2 SAFETY WARNINGS

- **CAUTION: SHOCK HAZARD** Although equipped with a safety interlock system, this apparatus should always be operated with extreme caution. Careless handling could result in electrical shock. The power supply should have open-circuit sensing.
- This apparatus should always be operated with caution. Careless handling can result in electrical shock.
- The system should be operated by trained personnel only.
- Some reagents indicated for use in this manual may be hazardous (*e.g.*, ethidium bromide, acetic acid, and boric acid, etc.); exercise care with these reagents.
- Always follow the power supply manufacturer's recommendations for use and follow safety procedures.
- Always turn off the DC power source *before* disconnecting the power cords from the apparatus.
- Never operate damaged or leaking equipment. Inspect the apparatus, electrical connections and power cords prior to use.
- For maximum safety, always operate this apparatus in an area that is not accessible to unauthorized personnel.

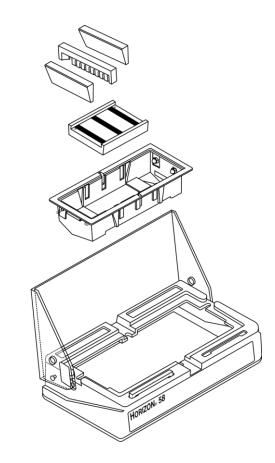
1.3 COMPONENTS

Components of the Horizon 58 Horizontal Gel Electrophoresis Apparatus are listed below:

- One tray support stand
- One formed polycarbonate buffer tray with multiple positioning slots for well-forming combs and integral Pt/Nb electrodes
- One machined acrylic gel deck
- One 3 and 4 stripe sample visualization loading guide
- Pair of aluminum gel casting dams
- Two precision machined Delrin[®] well-forming comb:
 - 8 tooth, 0.8 mm thick
 - o 14 tooth, 0.8 mm thick
- One pair 122 cm (48") red and black power cords
- One instruction manual

Figure 1. Horizon 58

The H58 Horizontal Gel Electrophoresis Apparatus (Figure 1) provides a compact and efficient unit for simplified gel casting and electrophoresis. It has been engineered for durable performance and easy storage. Many of these components are also available separately.



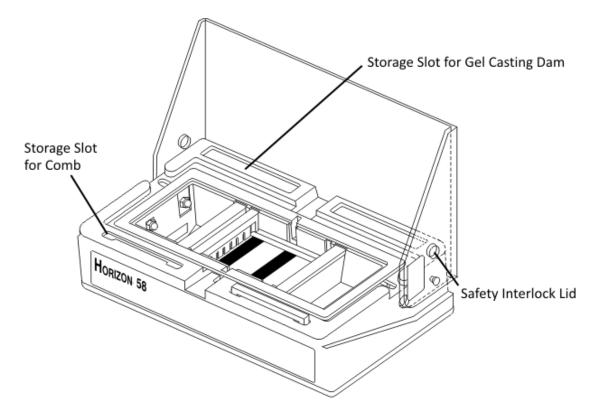


Figure 2. Tray Support Stand - The acrylonitrile butadiene styrene (ABS) plastic tray support stand features a clear acrylic safety interlock lid and storage facilities for the combs and gel casting dams.

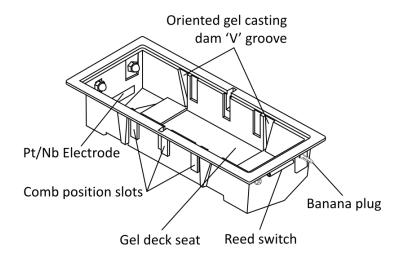


Figure 3. Buffer Tray - The clear polycarbonate plastic buffer tray is designed to be used in the tray support stand and not as a stand-alone electrophoresis apparatus. Because the buffer tray is removable, the electrophoresis buffer is easily discarded. The buffer tray provides three slots to position the combs and two 'V' grooves for the gel casting dams. Electrical contact is disabled when the buffer tray is free from the tray support stand due to the presence of reed switches on the outside ends of the tray.

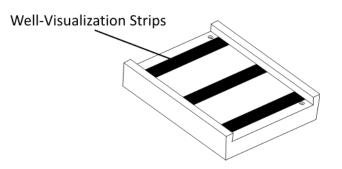


Figure 4. Gel Deck - The gel deck provides for a 5.7 x 8.3 cm gel bed and is designed to withstand the high temperatures of molten agarose without warping. Hence, gels may be cast immediately after boiling the agarose solution. Red well visualization stripes are provided by a card that is placed under the gel deck. There are four stripes on one side and three on the opposite side. Stripes are positioned under the comb(s) to permit easy viewing of the wells when loading a gel. The acrylic used in the gel deck is not UV transparent so gels should be removed from the gel deck for photography.

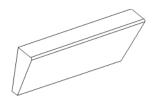


Figure 5. Gel Casting Dams - The gel casting dams are used to seal the ends of the gel deck when pouring agarose gels and eliminate the use of tape.

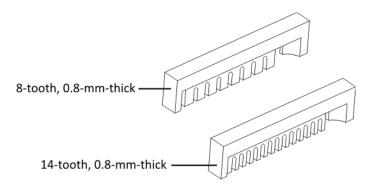


Figure 6. Combs - The white Delrin combs are used for casting wells in agarose gels. Two combs are standard components of the apparatus: one 8 tooth, 0.8 mm thick and one 14 tooth, 0.8 mm thick. (See Table 3 for capacities of wells generated by these combs.)

Power Cords - The 122 cm (48") long red and black safety shrouded power cords are used to connect the apparatus to a DC power supply.

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2.0 OPERATING INSTRUCTIONS

2.1 ASSEMBLY FOR GEL CASTING

Assemble the H58 apparatus on a level surface following this procedure. Refer to Figure 7 for the gel casting configuration of the components.

- 1. Open the safety interlocking lid and insert the buffer tray in the tray support stand. Do not cast or electrophorese gels with the buffer tray free from the tray support stand, as it is not a standalone unit.
- 2. Place the gel deck in the center of the buffer tray with the outermost well visualization strip towards the left (negative electrode).
- 3. Slide the gel casting dams down into the "V" grooves of the buffer tray. Apply gentle pressure simultaneously to both gel casting dams to seat the sealing surface against the sides of the gel deck. Do not force the gel casting dams down, as this may displace the gel deck out of level.
- 4. Insert the comb into the desired comb position slot, with the teeth in line with a well visualization strip on the gel deck. Multiple comb position slots are available for assorted comb placements (refer to Chapter 4, Applications). Ensure that the comb is resting unobstructed on the top of the gel deck.
- 5. Check that the surface of the gel deck is level with a circular 'bull's eye' level. Make adjustments if required.

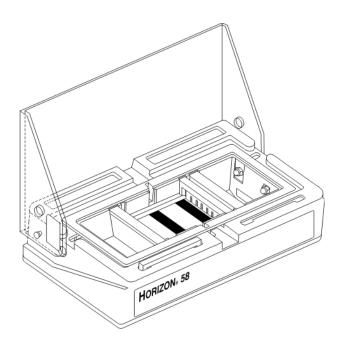


Figure 7, Gel Casting Configuration

2.2 PREPARING AGAROSE FOR GELS

To prepare a 1% (w/v) agarose gel, perform the following procedure:

- 1. Add 1 g of agarose to 100 ml of electrophoresis buffer in a 250 ml bottle or Erlenmeyer flask. Refer to Tables 1 and 2 for Tris acetate-EDTA (TAE) and Tris borate-EDTA (TBE) buffer formulas.
- 2. Loosely cap and weigh the flask.
- 3. Dissolve the agarose in electrophoresis buffer by heating in a microwave oven or boiling water bath with occasional mixing.
- 4. Weigh the flask and adjust the volume with deionized water to compensate for evaporation.

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Component	Amount	Concentration
Tris base	48.4 g	400 mM
Na ₂ EDTA•2H ₂ O	7.4 g	20 mM
Sodium acetate, anhydrous	16.4 g	200 mM
Glacial acetic acid	17.0 ml	296 mM
Deionized water	to 1 L	

Note: This is a 10X concentration solution. Dilute with deionized water prior to use. Final pH should be 7.8 at 25°C.

Table 2. 10X TBE Electrophoresis Buffer

Component	Amount	Concentration
Tris base	121.1 g	1 M
Boric acid, anhydrous	55.6 g	0.9 M
Na2EDTA•2H2O	3.7 g	10 mM
Deionized water	to 1 L	

Note: This is a 10X concentration solution. Dilute with deionized water prior to use. Final pH should be 8.3 at 25°C.

2.2.1 GEL CASTING PROCEDURE

To cast an agarose gel:

 Pipet the desired volume of molten agarose solution containing electrophoresis buffer into the apparatus assembled for gel casting. There is no need to cool the agarose to 50°C prior to casting the gel, as the gel deck can withstand high temperatures. Volumes required for various thickness gels are listed in Table 3.

WARNING: Do not cast gels with the buffer tray free from the tray support stand.

- 2. Ensure that the agarose is distributed evenly over the surface of the gel deck and remove any air bubbles.
- 3. Allow the agarose solution to cool until it solidifies.

For short term storage of gels:

- 1. Wet the surface of the gel with electrophoresis buffer.
- 2. Gently remove the comb and gel casting dams.
- 3. Lift the gel deck out of the buffer tray.
- 4. Wrap the gel in the gel deck with plastic wrap.
- 5. Store at 4°C (gels can be stored for 1 to 2 days).

Table 3. Sample Volumes for Horizon 58 Apparatus Combs as a Function of Gel Thickness

Gel Thickness (mm)	Agarose Volume (ml)	Comb Thickness (mm)	Number of Teeth	Capacity/Well (ul)	
		5	15		
		0.8	8	7	
			14	3	
3	15		3(b)	140	
5	15		5	30	
		1.5	8	15	
			14	7	
			5	20	
		8	10		
	0.8	14	5		
			3(b)	200	
4	20	1 5	5	40	
			8	20	
			1.5	12(c)	12
			14	10	
5 25		5	30		
		0.8	8	15	
			14	7	
		3(b)	250		
		1.5	5	50	
			8	25	
		14	13		

(a) Low percentage gels (<0.6%) and low melting point agarose gels may have lower sample volume.

- (b) Includes one preparative well and two flanking reference analytical wells for standards (dimensions and capacity values are for the central, preparative well).
- (c) 4.5 mm spacing

2.3 ELECTROPHORESIS

Add 1/10 volume of 10x sample loading buffer to samples. Apply directly to gel. Only samples containing cohesive ends (*e.g.*, lambda DNA restriction fragments) should be heated at 65°C for 10 min prior to loading. For additional sample loading buffers, refer to Maniatis *et al.* (1) or Rickwood and Hames (2).

Component	Amount	Final Concentration (10x)
Glycerol	5 ml	50% (v/v)
Na2EDTA•2H2O	0.37 g	100 mM
Sodium dodecyl sulfate	0.1 g	1% (v/v)
Bromophenol blue	0.01 g	0.1% (v/v)
Deionized water	to 10 ml	

Table 4. Sample Loading Buffer

To perform the electrophoresis procedure:

- 1. Remove the gel casting dams and return them to their storage slots in the tray support stand.
- 2. Pour 100 to 150 ml of electrophoresis buffer into the buffer tray. The surface of the gel should only be covered with 1 to 2 mm of electrophoresis buffer.
- 3. Gently remove the comb. To avoid tearing the bottom of the wells, gently wiggle the comb to free the teeth from the gel. Slightly lift up one side of the comb, then the other. Return the comb to its storage slot in the tray support stand.
- 4. Remove any trapped air bubbles to ensure that the wells fill with buffer.
- 5. Use a micropipette or automatic pipette to load the samples on the floor of the wells. Samples should contain sufficient glycerol or sucrose to be denser than the electrophoresis buffer. The formulation for a sample loading buffer is presented in Table 4. Loading capacities for each comb versus gel thickness are listed in Table 3.
- 6. Close the safety interlock lid.
- 7. Connect the power cords to the electrophoresis tank and a 250 VDC power supply. Connect the positive (red) lead at the right side of the apparatus and the negative (black) lead at the left.
- 8. Turn on the power supply and select the desired voltage. Small bubbles will rise from the electrodes when the unit is properly connected. Nominal electrophoresis times for TAE and TBE buffers are listed in Table 5.

Caution: Do not exceed the recommended voltage of the power supply.

Note: Nucleic acids will migrate toward the positive electrodes (red) at the right side of the apparatus.

 Monitor electrophoresis by following the migration of the bromophenol blue (BPB) dye. Movement should be in the direction of the positive electrodes at the right side of the apparatus. Use the black 1 cm graduations visible below the UVT tray to determine approximate migration rates. 10. When electrophoresis is complete, turn off the power supply. Disconnect the power cords from the power supply and then the apparatus.

Voltage (V)	Buffer (b)	Electrophoresis Time (c)
25	TBE	5 hours
25	TAE	5 hours
50	TBE	2.25 hours
50	TAE	2.25 hours
75	TBE	1.5 hours
/5	TAE	1.4 hours
100	TBE	60 min
100	TAE	56 min
125	TBE	45 min
125	TAE	42 min
150	TBE	36 min
150	TAE	34 min
475	TBE	29 min
175	TAE	27 min
200	TBE	24 min
200	TAE	22 min

 Table 5.
 Nominal Electrophoresis Time for 1% Agarose Gels at Constant Voltage (a)

- (a) Measurements were made with the gel submerged 1 to 2 mm and at normal operating current ranging from 5 to 125 mA.
- (b) Formulations for TAE and TBE electrophoresis buffers are listed in Tables 1 and 2.
- (c) Time required for BPB dye to migrate 6.5 cm from the origin. For a 1% gel, BPB co-migrates with DNA fragments of approximately 200 bp in 1x TBE and 400 bp in 1x TAE.

Caution: Electrophoresis >175 V generates sufficient heat to melt agarose gels. Do not exceed the high voltage electrophoresis times listed in Table 5 without cooling the gel during electro-phoresis. Do not run low melting point agarose gels at high voltages.

Note: Current and electrophoresis time vary with the volume of buffer, gel thickness, and voltage applied.

2.3.1 POST-ELECTROPHORESIS

After disconnecting the Horizon 58 apparatus from the power supply:

- 1. Open the safety interlock lid and lift the gel deck from the buffer tray.
- 2. Slide the gel out of the gel deck for staining or subsequent analysis (see Chapter 4, Applications). Remove the gel with care as agarose tears if not properly supported.
- 3. Remove the buffer tray from the tray support stand and properly discard the electrophoresis buffer. Do not reuse the buffer.

- 4. Rinse the buffer tray with deionized water.
- 5. Remove any residual agarose from the gel deck, gel casting dams, and combs by rinsing with deionized water. Wipe dry or allow to air dry before storing.
- 6. Store all components in the tray support stand.

3.0 TROUBLESHOOTING GUIDE

Some suggestions for resolving common problems are given below. Should these suggestions not resolve the problem, please call Technical Support (see Section 6.3 for numbers). If the unit must be returned for repair, also contact our service department, the technical support or your local distributor for shipping instructions. Please include a full description of the problem.

PROBLEM	COMMENTS
Bubbles do not appear on the electrodes when DC voltage is connected.	Verify that the DC power supply is operating properly. Verify continuity of the power cords with an ohmmeter.
Electrodes turn gray.	This occurs under normal operating conditions. Performance is not affected.
Agarose solution leaks during casting. the gel casting dams are clean.	Verify that the sealing surfaces of the UVT tray and Verify that the gel casting dams are properly seated.
BPB dye turns yellow (pH change) during electrophoresis. Results are uninterpretable.	Check the pH of the electrophoresis buffer (refer to Tables 1 and 2). Be sure to use Tris Base and not Tris-HCl. Mix the buffer periodically during electrophoresis.
Samples leak underneath the gel upon loading.	The bottom of the wells were torn when the comb was removed.
Pronounced 'smiling' along one edge of the gel occurs (corresponding bands in different lanes migrate slower toward one edge).	Gel was cast or electrophoresed out of level. Use the 'bull's eye' level to verify that the gel deck is level prior to gel casting and electrophoresis.
S-shaped lanes (anomalous migration-front results in lanes that are not all running at a uniform speed).	Mix the buffer periodically during electrophoresis. Switch to a low conductivity/high buffering capacity buffer (0.5X TBE). Reduce the salt concentration of the sample.
'Flaming' bands (excessive fluorescence appearing as a trail above the band)	Reduce the amount of DNA in the sample. Reduce the amount of protein and/or glycerol in the sample.
'Wiggly' or 'slanting' bands (bands are not straight lines or parallel to the top edges of the gel).	Verify that the wells are free of particles and bubbles before and after loading samples. Verify that the agarose is completely dissolved before casting gels. Remove any particulate matter from the agarose before casting gels. Be sure that bubbles are not trapped against the comb during gel casting.

All bands appear as 'doublets' (each band is represented twice within the same lane).

Concentrate the sample and use a thin (2 to 3 mm) gel with (0.8 mm) wells. Prevent gel movement during photography.

Reduce voltage. Band doublets may result as a result of denaturation from excess heat from running the gel at high voltage.

4.0 APPLICATIONS

4.1 CONSIDERATIONS FOR AGAROSE GEL ELECTROPHORESIS

4.1.1 SELECTING GEL CONCENTRATION

The selection of a percentage of agarose in a gel depends on the range of fragment sizes to be separated. Typically, 0.3% to 2% agarose gels are used. Large DNA fragments require low percentage gels, while small DNA fragments resolve on high percentage gels. Gels containing less than 0.5% agarose are very weak and electrophoresis should be performed at a low temperature (4°C). For routine electrophoresis, 0.75% to 1% agarose gels provide a wide range of separation (0.15 to 10 kb). A more complete treatment of factors that affect the separation of nucleic acids in agarose gels may be found in Maniatis *et al.* (1) or Rickwood and Hames (2).

The sample loading capacities that can be loaded per well for each available comb are listed in Table 3. For analytical purposes, the sample volume should be kept to a minimum. Generally, 0.8 mm wells provide sharper band definition than 1.5 mm wells.

Thin (2 to 3 mm thick) and low-percentage agarose gels yield better photographs than thick or high-percentage gels, which exhibit increased opaqueness and auto-fluorescence.

4.1.2 PREPARING SAMPLES AND LOADING THE GEL

The amount of DNA that can be loaded per well is variable and depends upon the number and size of the DNA fragments and the cross-sectional area of the well. As a general rule, the minimum amount of DNA detectable by ethidium bromide staining is 1 ng in a 5 mm wide band. For preparative purposes, 50 ng per 5 mm wide band should not be exceeded. Overloading the gel may cause trailing and distortion of the bands.

The multiple comb slots in the H58 apparatus lend themselves to a variety of applications. Two rows of wells increase the sample capacity of the gel for rapid screening of 'mini-prep' plasmids. A row of wells at the bottom of the gel is convenient to load quantitative standards for Southern blot hybridization just

4.1.3 ETHIDIUM BROMIDE STAINING OF DOUBLE-STRANDED DNA

To visualize double-stranded DNA after electrophoresis, the gel should be transferred from the gel tray to a 0.5 μ g/ml solution of ethidium bromide in deionized water. Approximate staining time is 10 to 15 min for a 3 mm thick gel and longer for thicker gels. As an optional subsequent step to reduce background fluorescence, the gel can be destained in deionized water for 15 to 30 min.

Alternatively, ethidium bromide may be added directly to the agarose prior to casting, so that the gel is electrophoresed in the presence of ethidium bromide. However, this procedure reduces the migration rate and may alter the relative electrophoretic mobility of nucleic acids (reference 3).

4.2 GEL PHOTOGRAPHY

A dark room or light-tight enclosure, film camera, digital camera and UV light source are required for photography of ethidium bromide stained gels. The stained gel is placed directly on top of a 300 nm transilluminator and photographed for a few second exposure at maximum aperture (f 4.5) with an appropriate film (e.g., Polaroid Type 57, ASA 3000). The intensity of the light source, the distance between the gel and the camera lens, the film speed, lens aperture, and the choice of photographic filters determine the exact exposure.

Note: The gel deck is not UV transparent and gels must be removed for photography.

Highest sensitivities (1 ng DNA in a 5 mm wide band) are obtained by photographing the gels with transmitted UV light. A UV blocking filter (e.g., Kodak[®] 2B Wratten filter), used in conjunction with a red gelatin filter (e.g., Kodak 23A Wratten filter), provides the highest contrast. Due to the fluorescence of the 2B filter, it is imperative that the two filters are oriented such that the red 23A filter is adjacent to the camera lens. The ethidium bromide-DNA complex fluoresces at 590 nm upon excitation at 302 nm (2). Short wave (254 nm) sources provide an equivalent level of sensitivity, however photodimerization and nicking of the DNA occurs. Long wave transilluminators (366 nm) are much less efficient. Photography under incident UV light is approximately 10-fold less sensitive than transmitted UV.

4.3 CONSIDERATIONS FOR ELECTROPHORESIS BUFFERS

For electrophoresis of agarose gels of the same concentration and at a fixed voltage, TAE buffer provides better resolution of high molecular weight fragments (>4 kb), while TBE buffer offers better low molecular weight resolution (0.1 to 3 kb). TBE has a higher buffering capacity and lower conductivity than TAE buffer. Hence, TBE is better suited for high voltage (>150 V) electrophoresis than TAE. TBE generates less heat at an equivalent voltage and maintains a constant pH.

Band compression of high molecular weight fragments (>5 kb) occurs as voltage increases. This effect is observed in both TBE and TAE buffers. Band definition remains sharp, even in excess of 200 V providing the gel is not overloaded. Linear DNA fragments from 0.15 to 10 kb (25 ng total) are easily resolved on a 0.8% agarose gel in 0.5x TBE run for 15 min at 200 V.

Electrophoresis at high voltages generates heat. High conductivity buffers, such as TAE, generate more heat than low conductivity buffers. Caution should be exercised in electrophoresis of gels >175 V. The heat build-up can cause gel artifacts such as 'S' shaped migration fronts, and prolonged electrophoresis can melt the agarose gel. Low melting point agarose gels should not be electrophoresed at high voltages. Nominal electrophoresis times for agarose gels in 1x TBE and TAE buffers at various voltages are listed in Table 5.

For the analysis of supercoiled DNA, TAE buffer produces better results. Anomalous migration of supercoiled DNA occurs when electrophoresed in TBE buffer >75 V. This effect is particularly pronounced with high molecular weight supercoiled DNA (>7 kb). The ability to resolve supercoiled DNAs from nicked circular and linear DNAs in the absence of ethidium bromide is also reduced. In order to accurately size supercoiled DNA, it is essential to electrophorese a standard in an adjacent lane (3).

The surface of the gel should only be covered with 1 to 2 mm of electrophoresis buffer. This prevents drying of the gel and ensures an even voltage gradient across the gel. Submerging the gel deeper than 2 mm is not necessary and results in increased current.

REFERENCES

- 1. Maniatis, T., Fritsch, E.F., and Sambrook, J. (1982) *Molecular Cloning: A Laboratory Manual,* Cold Spring Harbor Laboratory, Cold Spring Harbor, New York.
- 2. Rickwood, D. and Hames, B.D. (eds.) (1982) *Gel Electrophoresis of Nucleic Acids: A Practical Approach*, IRL Press, Oxford, England.
- 3. Longo, M.C. and Hartley, J.L. (1986) LTI Focus® 8:3.

5.0 RELATED PRODUCTS AND REPLACEMENT PARTS

5.1 H58 ACCESSORIES	DESCRIPTION	CATALOG #
Preparative precision machined white Delrin comb with marker lanes	1.5 mm thick	21065131
5 well precision machined white Delrin comb	0.8 mm thick 1.5 mm thick	21065099 21065107
8 well precision machined white Delrin comb	0.8 mm thick 1.5 mm thick	21065073 21065081
14 well precision machined white Delrin comb	0.8 mm thick 1.5 mm thick	21065115 21065123
12 well precision machined white Delrin comb	4.5 mm spacing	11951142
Buffer Tray, Assembled with both electrodes	Each	21065024
H58 Gel Deck machined acrylic, <u>includes</u> card below 3 and 4 stripe sample loading visualization card	Each Replacement card	21065164 21065165
H58 Aluminum Casting Dams	Package of 2	21065065
Power Cord Replacements (high voltage) 1 black and 1 red, 122 cm (48") long	Package of 2	11099025
H58 Pt/Nb Electrode Replacement (1 electrode) Includes all necessary components	Kit	21059027

6.0 CARE AND HANDLING

6.1 MATERIALS AND CARE

The H58 apparatus and related accessories are fabricated from ABS, polycarbonate, Delrin, and acrylic plastics. As with any laboratory instrument, adequate care yields consistent and reliable performance. Acrylic and ABS both have very good heat, impact, and chemical resistance but will not withstand autoclaving.

Caution: Both electrodes are made from Pt/Nb strip for durability but use care when cleaning this apparatus to prevent breakage of the electrodes because they <u>are not</u> warranted against breakage.

All components may be washed with water and a detergent. To remove grease and oils, use a hexane, kerosene, or aliphatic naphtha. *Never* use abrasive cleaners, window sprays, or any fluid that may contain toluene, methylene chloride, phenol, acetone, benzene, halogenated hydrocarbon solvents, or undiluted laboratory alcohols. Avoid prolonged exposure of the apparatus to UV light.

Routine inspection and maintenance will ensure both the safety and the performance of your horizontal gel apparatus. For replacement parts, call your distributor or Apogee Technical Support.

- Because of the relatively high voltages that may be used, inspect electrical connections and power cords often. If power cords show any signs of wear or damage (e.g., cracks, nicks, abrasions, melted insulation or bare wire), replace immediately.
- Examine the electrode banana plugs and connection nuts to ensure that they are free of corrosion or they may offer higher resistance thus heating up and risking sparks and fire.

Specification	Horizon 58
Weight	0.64 kg (1.13 lb)
Dimensions (W × L x H)	15.2 x 24.0 x 7.0 cm (6.0 x 9.4 x 2.8 in)
Gel Dimensions	5.7 x 8.3 x 0.05 cm (2.25 x 3.25 x 0.2 in)
Maximum gel thickness	10 mm
Buffer volume	100 to 150 ml
Voltage Range	200 VDC Max
Current Range	4 – 360 mA, 0.5 Max
Electrode material	Pt/Nb strip
Operating Temperature Range	4-30°C
Construction	ABS, acrylic, polycarbonate, Delrin, aluminum

6.2 GENERAL SPECIFICATIONS

6.3 TECHNICAL SUPPORT AND SERVICE

Should you have any problems with this unit, please contact:

Apogee Designs, Ltd. Attn: Electrophoresis Support 101 Kane Street Baltimore, MD 21224 USA

Phone: 443.744.0368 9 to 5PM EST, Monday through Friday Fax: 410.633.3666 Email: info@apogeephoresis.com

6.4 INSTRUCTIONS FOR RETURN SHIPMENT

IMPORTANT: Before sending the unit back to us, it is absolutely necessary to call our Technical Support department to **get authorization to return products**!

- Return only defective devices. For technical problems which are not definitively recognizable as device faults please contact Apogee Technical Support.
- Use the original box or a similarly sturdy one.
- Label the outside of the box with CAUTION! SENSITIVE INSTRUMENT!
- Please enclose a detailed description of the fault and when, or how, the problem occurred.

Important: Clean all parts of the instrument from residues and of biologically dangerous, chemical and radioactive contaminants. Please include a written confirmation (use the respective Decontamination Declaration/Certificate following in Section 8 that the device is free of biologically dangerous and radioactive contaminants in each shipment. If the device is contaminated, it is possible that Apogee will be forced to refuse to accept the device. The sender of the repair order will be held liable for possible damages resulting from insufficient decontamination of the device.

Please enclose a note which contains the following:

- 1. Sender's name and address and,
- 2. Name of a contact person for further inquiries with telephone number.

6.4.1 CLEANING AND DECONTAMINATION FOR RETURN OF PRODUCTS

Use the original product packaging whenever possible, to avoid damage to the unit being returned. All returned material must be cleaned and decontaminated prior to shipping. The components of apparatus products are fabricated from a variety of materials including: ABS, acrylic, vinyl, glass, silicone, aluminum and stainless steel.

Please clean any unit or product to be returned using the following three step procedure.

STEP 1: GENERAL CLEANING PROCEDURE

For materials not contaminated with biological or radiological substances, components may be gently washed with water and a non-abrasive detergent, and rinsed with deionized water. Dry using a soft cloth, paper towel or allow to air dry. A light application of hexane, kerosene, or aliphatic naphtha will remove grease.

To prevent surface damage, never use abrasive cleaners, window sprays or scouring pads to clean these products. Avoid excessive exposure to UV light, phenol, acetone, benzene, halogenated hydrocarbon solvents or undiluted alcohols because they may cause crazing.

STEP 2: BIOLOGICAL CLEANING PROCEDURE

Using a solution of either 5% household bleach in water or 70% ethanol in water, wipe down the apparatus using a clean cloth or sponge. Neutralize the solution by wiping the surface with a mild, nonabrasive detergent and rinse well with water.

STEP 3: RADIOLOGICAL DECONTAMINATION PROCEDURE

To meet various regulatory and safety standards, please follow the decontamination procedure given here if radioactive materials are used with this product or are used in the vicinity of where this apparatus has been used or stored.

WARNING: We cannot and will not accept return of products that are contaminated with any radioactivity.

For beta emitting isotopes such as ³²P, use a GM-type radioactivity meter calibrated in counts per minute (CPM) to determine the background readings for your work area. Wearing latex gloves, survey the unit to be returned with the GM meter. If any part of the unit is found to show readings higher than background, wash the area using Radiacwash[®] (Atomic Products Corp.) and paper towels, or another similar commercially available detergent. If none are available use a mild detergent or Formula 409[®] like solution. As you clean, discard liquid and solid waste (gloves and paper towels) according to your local and institutional regulations for radioactive material disposal. Continue washing until the GM-meter reading for the contaminated area(s) is equal to or below background.

To decontaminate units where a GM-meter is not as useful for detection, as with 'H, or "S, it will be necessary to perform swipes of the unit and detect using a scintillation counter. The unit should be dry. Wipe surfaces with dry paper circles (these are commercially available or you can make your own). Areas can be charted, so that individual swipes can be done on different surfaces to better isolate areas of contamination.

Swipes should be placed into individual scintillation vials with an appropriate floor and then analyzed on a properly programmed scintillation counter. If contamination above 100 disintegrations per minute dpm/100cm² (dpm=CPM/efficiency) is found, wash the area as described above in ³²P decontamination. After cleaning the area, swipe it a second time to determine the amount of contamination remaining. If the area still has greater than 100 dpm/cm², continue the cycle of swipes and washing until you achieve a reading of less than 100 dpm/cm².

Once the unit has been determined to be radiation free (<100dpm/cm²) remove all the hazardous and radioactive labels from the unit. If the labels cannot be removed, deface them. Failure to do so may result in a significant delay or refusal of repair. If your unit has non removable contamination (detectable with a GM-meter and not with paper swipes, or detectable with paper swipes but after continued washing the dpm/cm² remains constant and above 100) of a short half life isotope such as ³²P, it may be stored for ten half lives of isotopic decay and the decontamination procedure repeated.

Note: Units contaminated with non removable, long half life isotopes may not be returned.

If questions still persist, please contact:

Apogee Designs, Ltd. Attn: Electrophoresis Support 101 Kane Street Baltimore, MD 21224 USA Phone: 443.744.0368 9 to 5PM EST, Monday through Friday Fax: 410.633.3666 Email: info@apogeephoresis.com

6.4.2 NOTICE REGARDING THE RETURN OF APPARATUS PRODUCTS

US Federal Regulations

In order to comply with US federal regulations and to protect the health and safety of employees, it is imperative that all customers read this notice and adhere to the requirements regarding the return of apparatus products. The US Department of Transportation, the Department of Health and Human Services, and the Nuclear Regulatory Commission have strict regulations on the shipment of hazardous materials (49 CFR Part 173) including etiologic agents (49 CFR Part 173 and 42 CFR Part 72) and radioactive materials (CFR 49 Part 173 and 10 CFR Part 20).

German Law

To comply with German law (i.e. §71 StrlSchV, §17 GefStoffV and §19 ChemG) and to avoid exposure to hazardous materials during handling or repair, completion of this form is required before equipment leaves your laboratory. When equipment is returned for repair, evaluation, credit or exchange, the <u>customer becomes the shipper</u> and must ensure that the item is free of contamination whether chemical, biological or radioactive. Procedures for decontamination are described above.

Materials received that have not been properly decontaminated or units which do not have hazard labels (such as 'caution radioactive materials') may be decontaminated at the customer's expense (approximately \$350) and may result in delay or refusal of repair. In addition, in the case of radioactive contamination, Apogee may be required to notify a licensing authority who in turn may be required to notify the customer's licensing authority.

Please carefully follow the instructions on decontamination and fill out the Decontamination Declaration that follows. Place the Decontamination Declaration inside the top flap of the box where it can be immediately noticed by the receiver. Any change to this procedure may result in service delay.

7.0 WARRANTY

7.1 WARRANTY

Apogee warrants apparatus of its manufacture against defects in materials and workmanship, under normal service, for one year from the date of receipt by the purchaser. This warranty excludes damages resulting from shipping, misuse, carelessness, or neglect and does not include breakage of the electrodes or crazing from cleaning with solvents that attack ABS or acrylic. Apogee's liability under the warranty is limited to the repair of such defects or the replacement of the product, at its option, and is subject to receipt of reasonable proof by the customer that the defect is embraced within the terms of the warranty. All claims made under this warranty must be presented to within three years following the date of delivery of the product to the customer.

This warranty is in lieu of any other warranties or guarantees, expressed or implied, arising by law or otherwise. Apogee makes no other warranty, expressed or implied, including warranties of merchantability or fitness for a particular purpose. Under no circumstances shall Apogee be liable for damages either consequential, compensatory, incidental or special, sounding in negligence, strict liability, breach of warranty or any other theory, arising out of the use of the product listed herein.

In the interest of bettering performance, Apogee reserves the right to make improvements to the design, construction, and appearance without notice.

7.2 DECLARATION OF CONFORMITY AND CE MARK

Note: The information outlined in this section applies only to customers located in the European Union (EU).

This laboratory apparatus is identified with the CE mark. This mark indicates that the product complies with the following EU Directives and Standards:

APPLICATION OF COUNCIL DIRECTIVE(S):

89/336/EEC	Electromagnetic Compatibility
73/23/EEC	Low Voltage Directive
STANDARDS:	
EN 50081-1:1992	Emissions
EN 50082-1:1992	Immunity
EN 61010-1:1993	Product Safety

8.0 DECONTAMINATION DECLARATION

RGA Number (IMPORTANT):	
Customer Name:	
Institute:	
Address:	
TEL #:	FAX #:
E-mail:	
Unit type:	Serial number:
DESCRIPTION OF PROCEDURES USED TO DECON	TAMINATE UNIT (LOOK AT6.4.1)
1. Gently washed with water and a non-abrasive	e detergent, and rinsed with deionized water.
 2. Using a solution of 5% household bleach in w down using a clean cloth or sponge and neutron 	ater or 70% ethanol in water, the unit was wiped ralized with deionized water.
3. To meet various regulatory and safety standar given in 6.4.1 if radioactive materials were us	rds, please follow the decontamination procedures sed with this product.
This piece of equipment <u>has not</u> been decontamin	ated. Reason:
To the best of my knowledge, unit is free of che	mical, biological, or radioactive contamination.
I understand that if the equipment is found to be condocument, the equipment may be decontaminated a contaminated, the response time for repairs will be	at my expense. Also, if the equipment is found to be
Signature:	
Title:	
Date:	
Please place completed and signed form insi- immediately be noticed by the receiver. We the necessary precautions to ensure that handled by our employees.	appreciate you taking the time to perform