

Ordering Information

Catalog #	Description
K20-00001	ProteinChip H50 Buffer , 200 ml
K20-00005	ProteinChip H50 Buffer , 1 L
C57-30065	ProteinChip H50 Arrays , A-H format, 12
C50-30011	ProteinChip Cassette-Compatible Bioprocessor , includes ProteinChip array forceps, cassette hold-down frame, 12 blank ProteinChip arrays
C30-00001	ProteinChip CHCA Energy Absorbing Molecules (EAMs) , 5 mg/vial, 20
C30-00002	ProteinChip SPA Energy Absorbing Molecules (EAMs) , 5 mg/vial, 20
C30-00003	ProteinChip EAM-1 Energy Absorbing Molecules (EAMs) , 5 mg/vial, 20

The SELDI process is covered by US patents 5,719,060, 6,225,047, 6,579,719, and 6,818,411 and other issued patents and pending applications in the US and other jurisdictions.

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ProteinChip® H50 Buffer

Instruction Manual

Catalog #K20-00001

For technical support,
call your local Bio-Rad office, or
in the US, call **1-800-4BIORAD**
(1-800-424-6723).

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Uses

- Protein profiling and biomarker discovery
- Rapid protein analysis to determine purity, mass confirmation, or both

Introduction

ProteinChip H50 buffer is designed for use with the ProteinChip H50 array (catalog #C57-30065).

The ProteinChip H50 array surface binds proteins through reverse-phase or hydrophobic interaction chromatography and has binding characteristics similar to that of a C6 to C12 alkyl chromatographic resin.

In reverse-phase interactions, proteins within the sample are partitioned between the lipophilic phase of the array surface and the sample buffer. Proteins less hydrophobic relative to ProteinChip H50 binding buffer will not bind to the array surface, while proteins more hydrophobic will bind to the array surface. ProteinChip H50 buffer is a low-to-moderate-stringency buffer to allow maximal binding of proteins to the surface.

Storage

Store ProteinChip H50 buffer at 2–8°C.

Buffer Composition

10% acetonitrile, 0.1% trifluoroacetic acid, antimicrobial preservatives, 200 ml — Amount is sufficient to run 12 ProteinChip H50 arrays in a ProteinChip cassette-compatible bioprocessor (catalog #C50-30011) using binding and elution buffer volumes as outlined in the suggested protocol below.

Suggested Protocol

1. (Optional) Bulk-wash the ProteinChip arrays with 50% methanol or acetonitrile for five minutes. Repeat once. Dry the arrays for 1 hour after bulk wash to minimize any spot-to-spot cross-contamination.
2. Assemble the ProteinChip arrays in the ProteinChip cassette-compatible bioprocessor.
3. Add 100 µl of ProteinChip H50 buffer to each well. Vortex for 5 minutes at room temperature.
4. Remove buffer from wells.

5. Add 90 µl of ProteinChip H50 buffer to each well.
6. Add 10 µl of sample to each well. Vortex for 30 minutes at room temperature.
7. Remove samples from wells.
8. Wash each well with 100 µl ProteinChip H50 buffer for 5 minutes, with agitation. Repeat once for a total of two buffer washes.
9. Remove wash buffer from wells and rinse each well with deionized water.
10. Drain wells and remove arrays from the ProteinChip bioprocessor.
11. Allow arrays to air-dry.
12. Apply 1.0 µl ProteinChip energy absorbing molecule (EAM) solution per spot. Two applications of EAM solution can be used in order to increase signal intensity. Allow arrays to air-dry.
13. Analyze arrays using the ProteinChip SELDI reader.