SPRI Automated gDNA Extraction from iSWABs

Jan 2014
iSwab from Mawi DNA Technologies

- Buccal swabs
- Comb in the cap of the collection tube, so cells are scraped off into a preservative
- No need to squeeze or clip swabs

– http://mawidna.com/
SPRI Extraction of iSwabs

• Two Beckman Coulter gDNA extraction kits tested
  – DNAdvance (designed for mammalian tissue)
  – Genfind v2 (designed for whole blood)
• Biomek 4000 (B4k) automated methods used for both
• Only part not automatable is aliquoting sample to plates from the iSwab tube.
  – The comb necessitates manual pipetting with a p200 to get most of the sample
  – Everything else is straightforward
DNAdvance

- Automated 96MC method adapted for higher input volume to accommodate liquid sample (A48708)
- Per well:
  - 300uL Sample + 193uL Lysis + 7uL Proteinase K
- Digest at 37C for 1hr
- Run Settings

<table>
<thead>
<tr>
<th>Step</th>
<th>Lysate Vol</th>
<th>Bind Buf Vol</th>
<th>Bead Vol</th>
<th>Ethanol Vol</th>
<th>Elution Vol</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>500</td>
<td>200</td>
<td>340</td>
<td>700</td>
<td>50</td>
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<td>1</td>
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- Shake Sec: 60
- Inc Min: 1
- Cycles: 3
- Transfer Vol: 45
DNAAdvance Results

- Modified run settings and DNAAdvance 96MC method produces significant yields
- Single plate 1 hour
Genfind v2

- 300uL input
- Default Span8 Method (A42569)
  - Elution in 50uL
Genfind v2 Results

- Genfind v2 and default Span8 method successfully extract gDNA
- Lower Yields than DNAdvance, likely due to shorter lysis at Room Temperature
Comparison of the two Kits

- Both DNAdvance and Genfind v2 successfully extract gDNA from iSWABs
- Higher yield from DNAdvance likely due to lysis:
  - 37C 1hr vs Room Temperature 30min on deck
## Ordering Information

<table>
<thead>
<tr>
<th>Product</th>
<th>Part Number</th>
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<tbody>
<tr>
<td>DNAdvance</td>
<td>A48705 (384 preps), A48706 (9600 preps)</td>
</tr>
<tr>
<td>Genfind v2</td>
<td>A41499 (50 preps), A41497 (384 preps), A83078 (4800 preps)</td>
</tr>
<tr>
<td>Biomek 4000 Liquid Handler</td>
<td>B22867</td>
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</tbody>
</table>

www.beckmancoulter.com

• 260/280 consistently ~2.0
• 260/230 varies mainly by donor
Gel analysis—gDNA Size / Stability

- Full length gDNA
- gDNA smears at up to 40kb
- Samples from each of 5 donors
  - Eluted in DNAdvance/ or Genfind v2 elution buffer
  - Extracted gDNA was Stored at 4°C for 3 weeks
  - Run on 0.8% agarose gel