QTLs From Genome to Field Using Markers and Genetic Maps for Peanut Improvement

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Constraints for low production
Drought
Leaf spot
Leaf rust

Integrated genomics approaches in peanut

Marker series | Markers | References | Research Institute/University
--- | --- | --- | ---
Arachis | 24 | Papai et al. 2004 | University of Georgia, USA/Cornell University, USA
| 236 | Shu et al. 2004 | Universidade Estadual Paulista (UNESP), Brazil
| 32 | Hisoki et al. 2007 | USDA-ARS/Tuskegee University, USA
| 133 | Lee et al. 2008 | Universidade Federal Rural do Rio de Janeiro, Brazil
| 151 | Large et al. 2008 | USDA-ARS/Tuskegee University, USA
| 178 | Zhao et al. 2009 | Shandong Peanut Research Institute, China
| 290 | Wang et al. 2009 | USDA-ARS/Tuskegee University, USA
| 685 | Hwang et al. 2009 | University of Georgia, USA
| 113 | Lee et al. 2009 | USDA-ARS/Tuskegee University, USA
| 94 | Song et al. 2010 | University of Georgia, USA
| 210 | Liu et al. 2010 | USDA-ARS/Tuskegee University, USA
| 630 | Shi et al. 2010 | USDA-ARS/Tuskegee University, USA
| 102 | Shi et al. 2010 | University of Georgia, USA

Improved germplasm with resistance/tolerance to biotic/abiotic stresses; better nutritional quality and enhanced agronomic performance

~13,000 SSR markers

Contents

- Markers repository
- Genetic/consensus maps
- QTLs and linked markers
- Molecular breeding
- Summary

Arachis SSR markers

| Marker series | Markers | References | Research Institute/University
--- | --- | --- | ---
| AA, Ac, Ap | 149 | Pandey et al. 2002 | International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), India
| AA, Lec, Lep | 143 | Zhao et al. 2002 | Tuskegee University, USA
| AA, Lec, Lep | 63 | Macedo et al. 2012 | University of Brasilia, Brazil
| DArT features | 15,000 | Killian 2008, Varshney et al. 2012 | DArT Pty Ltd (Australia), ICRISAT (India), CIRAD (France), and Catholic University of Brasilia & EMBRAPA (Brazil)
| Single nucleotide polymorphism (SNP) markers | 1,536 | Ozias-Akins, Peggy, pers communication | University of Georgia, USA
| 768 | Cook, Douglas, pers communication | University of California-Davis, USA

DArT array showed low polymorphism

- Sequencing >200 genes in parents of mapping populations as well as screening of 1536 SNPs (from U6, USA) also yielded very few polymorphic SNPs
- As a collaborative effort with UGA, USA (Dr Peggy Ozias-Akins), a set of 96 highly informative SNPs in cultivated germplasm were converted into KASPar assays
- Validated KASPar assays available for 90 SNPs

Five genetic maps for 4x groundnut

<table>
<thead>
<tr>
<th>Details of different linkage maps</th>
<th>TAG 24</th>
<th>ICGS 76</th>
<th>ICGS 44</th>
</tr>
</thead>
<tbody>
<tr>
<td>IGV 86031</td>
<td>x</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>RIL-1 (RIL-2)</td>
<td>IGV 76</td>
<td>x</td>
<td>x</td>
</tr>
<tr>
<td>RIL-3 (RIL-4)</td>
<td>CSMG 84-1</td>
<td>x</td>
<td>6PBD 4</td>
</tr>
<tr>
<td>RIL-5 (RIL-6)</td>
<td>ICGS 76</td>
<td>x</td>
<td>6PBD 4</td>
</tr>
</tbody>
</table>

Marker loci mapped: 191

Linkage groups: 22

Marker loci/LG: 2-19

Avg. marker loci/LG: 8

Total map distance (cM): 1785

Avg. distance/LG (cM): 81.15

Avg. inter-Locus distance (cM): 9.54


Reference consensus genetic map

Marker loci mapped: 897

Total map distance: 3863.6 (cM)

Map density: 4.42 (cM)

PloS ONE 2012, 7 (7): e41213

Eleven 4X- genetic maps used for consensus map

<table>
<thead>
<tr>
<th>Maps</th>
<th>Linkage groups</th>
<th>Polymorphic loci</th>
<th>Mapped loci</th>
<th>Map length (cM)</th>
<th>Map density (cM)</th>
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</thead>
<tbody>
<tr>
<td>RIL-1</td>
<td>22</td>
<td>211</td>
<td>191</td>
<td>1785.4</td>
<td>9.35</td>
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<tr>
<td>RIL-2</td>
<td>20</td>
<td>128</td>
<td>119</td>
<td>2018.2</td>
<td>18.56</td>
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<tr>
<td>RIL-3</td>
<td>15</td>
<td>87</td>
<td>82</td>
<td>831.4</td>
<td>10.14</td>
</tr>
<tr>
<td>RIL-4</td>
<td>20</td>
<td>209</td>
<td>188</td>
<td>1922.4</td>
<td>10.23</td>
</tr>
<tr>
<td>RIL-5</td>
<td>21</td>
<td>209</td>
<td>181</td>
<td>1963</td>
<td>10.85</td>
</tr>
<tr>
<td>RIL-6</td>
<td>19</td>
<td>146</td>
<td>132</td>
<td>700.4</td>
<td>6.01</td>
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<tr>
<td>RIL-7</td>
<td>21</td>
<td>124</td>
<td>109</td>
<td>503.1</td>
<td>4.62</td>
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<tr>
<td>RIL-8</td>
<td>13</td>
<td>64</td>
<td>46</td>
<td>357.4</td>
<td>7.76</td>
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<tr>
<td>RIL-9</td>
<td>26</td>
<td>261</td>
<td>233</td>
<td>1304.9</td>
<td>5.6</td>
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<tr>
<td>RIL-10</td>
<td>22</td>
<td>183</td>
<td>175</td>
<td>917.45</td>
<td>5.3</td>
</tr>
<tr>
<td>BCF1-11</td>
<td>21</td>
<td>339</td>
<td>332</td>
<td>847.4</td>
<td>2.53</td>
</tr>
</tbody>
</table>

Genus map: 20 - 987 3863.6 4.42

Markers repository

- Genetic/consensus maps
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An International Reference Consensus Genetic Map with 897 Marker Loci Based on 11 Mapping Populations for Tetraptlate Groundnut (Arachis hypogaea L.)

Marker loci mapped: 3,693

Total map distance (cM): 2,651

Map density (loci/cM): 1.39

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Shirasa et al. 2015, DNA Research
16 QTL clusters identified for drought related traits

<table>
<thead>
<tr>
<th>Cluster</th>
<th>QTL Traits</th>
<th>R² (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cluster 1</td>
<td>LDW, TW, SORD, TORDER, WD, T1E</td>
<td>3.64-2.23</td>
</tr>
<tr>
<td>Cluster 2</td>
<td>SLA, SORD, TORDER</td>
<td>3.91-2.24</td>
</tr>
<tr>
<td>Cluster 3</td>
<td>SciWR, SciMD, TDW, Veg wt</td>
<td>5.06-3.36</td>
</tr>
<tr>
<td>Cluster 4</td>
<td>HD, TORDER</td>
<td>6.91-7.29</td>
</tr>
<tr>
<td>Cluster 5</td>
<td>TORDER, SORD, Pod wt, SORD, Haulm wt, TDW68125,</td>
<td>1.7-13.44</td>
</tr>
<tr>
<td>Cluster 6</td>
<td>LA, SORD, Pod wt, TDW, TORDER, SLA, SORD, SORD, TORDER, TE</td>
<td>2.93-9.85</td>
</tr>
<tr>
<td>Cluster 7</td>
<td>SLA, Haulm wt, SciWR, SciMD, TORDER</td>
<td>3.90-9.87</td>
</tr>
<tr>
<td>Cluster 8</td>
<td>SciWR, SciMD, LA</td>
<td>6.43-10.49</td>
</tr>
<tr>
<td>Cluster 9</td>
<td>SciWR, Pod wt, Haulm wt, LA, TORDER</td>
<td>7.30-12.15</td>
</tr>
<tr>
<td>Cluster 10</td>
<td>SciWR, Pod wt, Haulm wt, LA, TORDER</td>
<td>4.67-7.74</td>
</tr>
<tr>
<td>Cluster 11</td>
<td>Initial DW, SLAG, SORD, TORDER, Haulm wt, SciMD, SciWR, TE68124</td>
<td>4.19-20.32</td>
</tr>
<tr>
<td>Cluster 12</td>
<td>TORDER, Haulm wt, SORD, LA, SciWR, TORDER, SciWR</td>
<td>3.44-12.80</td>
</tr>
<tr>
<td>Cluster 13</td>
<td>SLAG, SORD, TORDER</td>
<td>3.5-13.95</td>
</tr>
<tr>
<td>Cluster 14</td>
<td>HD, Veg wt</td>
<td>6.62-40.30</td>
</tr>
<tr>
<td>Cluster 15</td>
<td>SciWR, SciMD, SORD</td>
<td>5.41-19.53</td>
</tr>
<tr>
<td>Cluster 16</td>
<td>Total DW, SciWR,</td>
<td>2.51-9.87</td>
</tr>
</tbody>
</table>

Mol Breeding 2012, 32:757-772
A major QTL for rust in peanut (PVE up to 82.96%)

QTLs/markers for molecular breeding

A diagnostic marker for rust resistance

Importance of high oleic acid in peanut oil

- Peanut seeds contain about 44-56% oil which comprised up to 12 fatty acids.
- Two major fatty acids oleic and linoleic account for approximately 80% of the oil composition (Ahmed and Young 1982).
- Peanut oil with high % of linoleic acid are prone to oxidation, leading to rancidity, off-flavors, and short shelf-life during seed storage.
- High levels of oleic acid is beneficial to human health by reducing low-density lipoproteins, maintaining high-density lipoprotein, slowing down atherosclerosis, and reversing the inhibitory effect of insulin production.

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Marker interval

- Oleic/linoleic acid ratio
- Oleic acid
- Linoleic acid
- LLS
- Rust

Marker interval

- Oleic acid 2010
- Oleic acid 2011
- Oleic/linoleic acid ratio

Marker interval

- Marker effects
- Marker interval
- Marker significance

Marker interval

- Marker significance
- Marker interval
- Marker effects

Marker interval

- Marker significance
- Marker interval
- Marker effects
Map saturation and new co-dominant markers

- Identification of four more markers (GM2009, GM2079, GM1536, GM2301), all four markers are co-dominant in nature as compared to dominant marker IPAHM103.

- All these markers are deployed to introgress QTL for rust resistance into the genetic background of three elite cultivars (TAG 24, JL 24 and ICGV 91114).

- This is the first effort towards moving QTL from one to another genotype through marker-assisted breeding to improve any trait in peanut.

Marker-assisted breeding for rust resistance

Phenotypic expression of rust QTL

Allele specific markers for oleate trait

Marker-assisted breeding for high oleate trait

Yield assessment of promising resistant lines
Summary

- Development of large number of markers
- Construction of several SSR based genetic linkage maps for cultivated groundnut
- Development of highly dense consensus genetic maps
- Genome sequencing is in progress for diploid and tetraploids
- Involvement of few main effect (M-QTLs) and several epistatic (E-QTLs) QTLs for drought tolerance related traits
- Availability of linked markers for foliar diseases (rust and late leaf spot), tomato spotted wilt virus (TSWV) and high oleate trait
- Initiation of marker-assisted introgression of foliar diseases and high oleate trait for improvement of elite cultivars
- Promising introgression lines possessing rust QTL available with improved resistance

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Thanks for your patience