Introduction

It's exciting to see how well a 1-stage workflow works... the data looks really good...

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Amp-Seq One simpler and more efficient targeted GBS

LGC Biosearch Technologies is excited to announce <u>Amp-Seq One</u>, our new 1-stage Amp-Seq technology for targeted genotyping by sequencing (GBS), including all the benefits of original Amp-Seq with only one PCR stage – half the labour, double the throughput.



Figure 1. Schematic of the Amp-Seq One Reagent System workflow. Total workflow time is 2 hours.

Amp-Seq One - key benefits

- **Simplicity** simplified workflow (see figure 1) with a reduced number of reagents resulting in half the hands-on time and reduced labour costs, minimising any risk of sample crosscontamination, as well as maximising possibilities for workflow automation.
- Throughput reduces the total time required to complete the library preparation by over 1 hour and doubles throughput versus traditional 2-stage methods.
- Cost eliminates the requirement for liquid handling equipment to transfer material between reaction stages. This results in

reduced capital expenses and maintenance costs as well as reduced consumable cost.

- **Density** targets thousands of markers in a single reaction (see figure 2)
- **Compatibility** typically compatible with 2-stage designs, common automation platforms, diverse sample materials and purification methods, diverse plant and animal species and multiple sequencing platforms – data to be published and not yet shown in this announcement.

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	Targeted GBS technology			
Protocol parameter	Amp-Seq One	Original Amp-Seq	Next fastest targeted GBS method	
Amplification time	50 minutes	100 minutes	180+ minutes	
Protocol turnaround time*	120 minutes	180 minutes	235+ minutes	
Sample transfer**	None	Required	Required	
Number of plates used per sample	1	2	2	

Table 1. Summary of protocol parameters for Amp-Seq One, original Amp-Seq and the next fastest amplicon-based targeted GBS method. *Turnaround time defined by processing one plate from starting materials to sequencing-ready library as prepared on standardly available liquid handling platforms. **referring to sample transfer from PCR 1 plate to PCR 2 plate

Amp-Seq One - performance

Amp-Seq One delivers high quality sequencing data on a wide variety of species, automation platforms and on diverse sample materials obtained from various purification methods. Here we present a dataset generated with Amp-Seq One. The dataset is based on a 1,920 marker panel for maize (B73, unmodified 2-stage panel) and demonstrates comparable performance between the original Amp-Seq protocol (requiring 2 PCR stages) and our new Amp-Seq One technology (figure 2).



Figure 2. Dataset for 1,920 marker panel for maize, illustrating comparable performance between Amp-Seq One and original Amp-Seq (2 PCR stages).

Sequencing data quality values are summarised in table 2. For both the original protocol and the Amp-Seq One protocol, percentage of reads mapped to the reference genome was greater than 98.5%, and the percentage of reads mapped to the target SNP location was greater than 95%. Genotyping call rates and uniformity (read depth consistency across markers) were also comparable between both methods.

	Percentage reads mapped	Percentage reads on target	Percentage of markers called	Uniformity
Amp-Seq One	98.7%	96.6%	96.9	89.1
Original Amp-Seq	99.4%	99.1%	96.7	88.9

Table 2. Sequencing data quality characteristics for maize samples using a 1,920 marker Amp-Seq panel, downsampled to an average depth of 200X. Data demonstrates high quality sequencing results for both Amp-Seq One and original Amp-Seq.

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Accelerate your breeding programme with Amp-Seq

Our <u>Amp-Seq technology</u> offers a high speed, high accuracy targeted NGS solution, bringing together the benefits of NGS with the ability to analyse thousands of samples in the same timeframe as end-point genotyping. This provides game-changing benefits to agricultural biotechnology companies as it increases throughput, resulting in higher predictive breeding accuracy and an accelerated time to market for new traits.

Capable of multiplexing thousands of markers in a single assay, Amp-Seq enables consolidation of screening strategies for genomic selection and marker-assisted selection, increases screening power of genotyping at key selection stages within breeding programmes, increases the accuracy and power of selection and limits data redundancy.

Panel design

We work with you to design a custom panel that meets your genotyping goals. With our proprietary design software, we design high-quality panels that result in high sequencing efficiency and typically have very high SNP call rates and evenness of coverage. These panels can be further refined and optimised for high priority markers.

Simplifying data analysis

The BiosearchCaller analysis pipeline is specifically designed for Amp-Seq and

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is available without license for Amp-Seq customers. The pipeline is universally applicable to multiple species and panels and can be adapted to suit your requirements.

Sequencing flexibility

Amp-Seq is compatible with customers' sequencing needs, with amplicons that can be designed for sequencing lengths ranging from 50-150+ bp and for single and paired-end sequencing protocols. This provides flexibility in cost, time and data processing.

Availability and further information

Amp-Seq One is commercially available for industrial scale processing in the first half of 2025.

Find out how you can accelerate your breeding programme with the revolutionary Amp-Seq One workflow in your laboratory. <u>To start your</u> <u>Amp-Seq One evaluation, contact us today</u>.



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