Aquaculture genotyping

Efficient genotyping of Atlantic salmon using whole cell lysates and SwiftArrayStudio Microarray Analyzer

In this application note, we show:

- Applied Biosystems™ Axiom™ Salmon Microarray and SwiftArrayStudio™ Microarray Analyzer provide robust genotypes from salmon samples.
- Thermo Scientific™ Kingfisher™ Flex and Applied Biosystems™ MagMAX™ CORE system can be used for automated genomic DNA extraction from salmon samples.
- Applied Biosystems™ AgriSeq™ Genomic DNA extraction kit provides whole cell lysate option for collecting salmon genotypes.
- Purified genomic DNA and whole cell lysates provide indistinguishable genotyping data from salmon samples.

Introduction

Genetic testing is an important practice in modern aquaculture, enabling producers to optimize traits such as growth rate, disease resistance, and environmental tolerance while maintaining population health. By using tools like SNP genotyping and genomic selection, hatcheries can manage breeding programs with far greater precision, reducing inbreeding and improving stock performance [1]. At the same time, genetic testing plays a critical role in conservation, particularly in monitoring the interactions between farmed and wild populations. When farmed fish escape—an issue documented in Atlantic salmon [2] — they can interbreed with wild conspecifics, introducing domesticated alleles that have been selected for captivity rather than survival in natural ecosystems [3]. This gene flow can reduce the fitness of wild populations by disrupting locally adapted gene complexes, lowering genetic diversity, and ultimately degrading the resilience of wild genotypes. Consequently, integrating genetic monitoring into aquaculture practices is important not only for production efficiency but also for safeguarding the genetic integrity of surrounding ecosystems [4].

The analysis of large numbers of loci important for aquaculture has been facilitated by the development of microarrays specific to Atlantic salmon. A high-density Affymetrix™ Axiom™ Custom Array (Affymetrix was acquired by Thermo Fisher Scientific in 2016) querying 132,033 polymorphic SNPs was designed and validated using wild and farmed salmon populations [5]. A later Affymetrix Axiom array was designed to account for

Figure 1. Workflow used in this study for collecting DNA and analyzing salmon samples using SwiftArray solution. In this application note, we show how both purified DNA and whole cell lysates, without DNA purification, can be analyzed.

North American, European and Chilean populations of farmed Atlantic salmon [6]. This microarray was used characterize population structure and linkage disequilibrium in three Chilean farmed Atlantic salmon populations of different geographic origins [7]. Microarrays have also been used to identify disease resistance loci [8-9] and to characterize the genetic response to rearing environment on growth [10-11].

In order for aquaculturists to design breeding schemes and select for the desired traits, they need a fast and efficient method for genotyping fish. To meet the growing demands for ease of use and speed in microarray analyses, Thermo Fisher Scientific recently developed the Applied Biosystems™ SwiftArrayStudio™ Microarray Analyzer and SwiftArray™ Assay. This solution consists of a DNA target preparation workflow that can be completed in a single workday, and an instrument that automates hybridization, washing and scanning without any in-process touchpoints. The SwiftArrayStudio analyzer can provide data on an Applied Biosystems™ Axiom™ array plate after an overnight hybridization and scan (for further information, see **SwiftArrayStudio webpage**). This assay and improved workflow reduce the sample-to-answer time from five days to two days, allowing researchers to gather and interpret genetic data to keep pace with their needs.

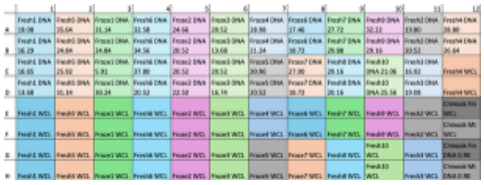
Description of experiments

In this application note, we show how the SwiftArrayStudio solution can be used for genotyping Atlantic salmon. Individual fresh or packaged frozen filet samples were purchased from three different local grocery stores, for a total of 17 samples. Approximately 5mm^3 replicate pieces were cut from each sample and processed as described below. Purified genomic DNA (gDNA) or crude whole cell lysate (WCL) samples were prepared for hybridization using the SwiftArray Assay kit according to the user guide. The targets were analyzed using the Applied Biosystems™ Axiom™ Salmon Genotyping Microarray for Atlantic salmon (*Salmo salar*), which queries around 130,000 variants found in Atlantic salmon, providing extensive information on polymorphisms across the salmon genome. Hybridization, washing and scanning was performed on the SwiftArrayStudio Microarray Analyzer using a 16 hr hybridization protocol. Subsequent analyses were performed using Axiom Analysis Suite 6.0 and best practices analysis parameters.

Sample extraction

To facilitate high-throughput preparation of samples, we used two methods that required minimal hands-on time. First, we used the Kingfisher™ Flex and the MagMAX™ CORE DNA

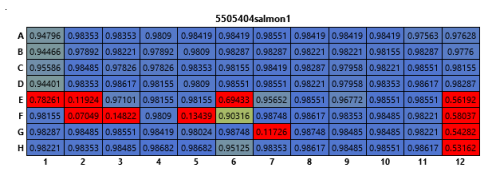
A.



B.

	1	2	3	4	5	6	7	8	9	10	11	12
A	1003	1086	1097	1065	1056	1062	996.0	1049	1016	961.7	817.3	858.5
B	929.1	992.2	1026	968.1	946.5	914.6	943.4	951.5	931.3	966.7	1026	991.5
C	913.7	1069	1076	1017	1040	1002	967.4	1029	1003	992.6	1072	1083
D	821.3	1044	1041	974.3	1000	978.4	921.3	1020	993.8	984.3	1044	1052
E	260.8	158.5	650.0	914.8	925.8	243.2	398.8	924.1	566.0	980.5	1027	1051
F	1017	230.8	310.7	1047	300.0	395.9	1076	1074	1043	1109	1047	1173
G	1081	1034	1050	991.5	940.1	986.2	194.4	928.4	938.4	1012	984.6	1109
H	1047	1006	1076	1037	1034	488.0	1083	999.3	1034	1010	1084	1108

C.



D.

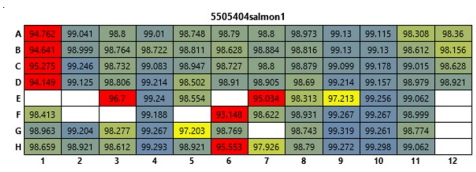


Figure 2. QC metrics of salmon samples analyzed. A. Layout of samples used. The lighter shades are samples that had gDNA purified and the amounts recovered; the darker shades are whole cell lysates (WCL). Note that samples taken from the same fish are colored similarly. Amount of DNA recovered is shown in micrograms. Amount of DNA in WCLs was not determined. B. Amount of DNA recovered after sample preparation using SwiftArray Assay workflow. Amounts are in micrograms; shading is from highest amount (green) to lowest amount (red). C. Dish QC values obtained from the samples, shaded from best (blue) to failing (red). D. QC Call rate for the samples. Note that samples that failed to meet minimal dQC values (0.82) were not evaluated by the Axiom Analysis Suite software (empty cells).

isolation kit to purify genomic DNA from the samples. Pieces were homogenized in 100µl of proteinase K buffer with blue pestle homogenizer, proteinase K added (10µl) and incubated at 65°C for 1 hour. Samples were spun and non-oil supernatant transferred to a fresh tube. 100µl of the supernatant was processed using the MagMax CORE kit, following the protocol recommended in the kit. Final DNA concentrations were determined using Invitrogen™ Qubit™ fluorometer. Total yield of genomic DNA averaged 2.3µg ± 0.6µg.

We also prepared crude whole cell lysates (WCL) for genotyping analysis using the Applied Biosystems™ AgriSeq™ Genomic DNA Extraction Kit. We homogenized the same approx. 5mm^3 tissue pieces in 100µl of the supplied lysis solution with blue homogenizer pestles. The homogenate was heated to 95°C for 10 minutes, followed by room temp for approx. 5min. An equal volume of stabilization solution was added, rehomogenized and debris spun out. Supernatant was transferred to a new tube.

Results of microarray analysis

After DNA extraction, the samples were processed using the SwiftArray Assay workflow. For purified gDNA samples, we used approximately 100ng of gDNA; for matched whole cell lysates, we used 50µl of supernatant (Figure 2A). Following the amplification, fragmentation and precipitation steps, the recovered. DNA was analyzed using the recommended quality control assays (**see SwiftArray Assay user guide here)**, including agarose gel electrophoresis (not shown) and quantification by absorbance at 260nm (Figure 2B). An average of 997.4 ± 66.7µg was produced from samples with purified genomic DNA. The whole cell lysates generated an average of 856 ± 308µg. Although the average amount recovered from lysates was slightly smaller, there was significant sample-to-sample variability, and in most cases there was enough to continue with data collection.

The samples were next loaded on the SwiftArrayStudio Microarray Analyzer for automated hybridization, washing and scanning. We analyzed the QC metrics generated by the Axiom Analysis Suite and default best practices parameters (see **Axiom Analysis Suite user guide here**). One of these, DishQC (DQC) measures the amount of overlap between two homozygous peaks created by non-polymorphic probes, with a value of 1.0 considered optimal. All of the purified gDNA samples (48/48) passed DQC above the best practices default threshold (0.82). For the WCL samples, 39/48 passed DQC; however, for each replicate, at least two lysate samples passed DQC (Figure 2C) and could be analyzed further.

Another quality measurement generated by Axiom Analysis Suite is the average passing QC call rate (average percentage of 200 autosomal SNPs with a call other than NoCall, as measured at the Sample QC step of passing samples within a plate).

Table 1. Marker metrics summary for salmon samples

	Purified gDNA		Whole Cell Lysates	
	Count	Percent	Count	Percent
NoMinorHom	49919	39.445	47460	37.502
PolyHighResolution	41907	33.114	38666	30.553
MonoHighResolution	21708	17.153	21698	17.145
Other	9509	7.514	10371	8.195
CallRateBelowThreshold	3344	2.642	8065	6.373
OTV	167	0.132	294	0.232

Among purified gDNA that passed DQC, six failed the default call rate threshold (97%). Interestingly, four of these were from one sample (A1-D1). The final two (G12 and H12) were from sockeye (Pacific salmon) and might not be expected to pass under default conditions. Among the WCL samples, in addition to the 9 that failed DQC, four more failed to pass call rate QC (Figure 2D). However, replicates from those samples were able to pass, suggesting the failure was not due to the ability to analyze lysates, rather due to a particular lysate instance.

To compare the data quality of the gDNA versus whole cell lysate samples, we analyzed the genotypic clustering quality (Table 1). Overall, the number of markers that fell into the different marker classes agreed very well. The biggest differences were in the less informative classes (Call Rate Below Threshold, Other, OTV). Finally, we compared the cluster plots of calls made from gDNA and WCL (Figure 3). A vast majority of the assigned calls were completely congruent (Figure 3A), even if some of the replicates failed to generate data (Figure 3B).

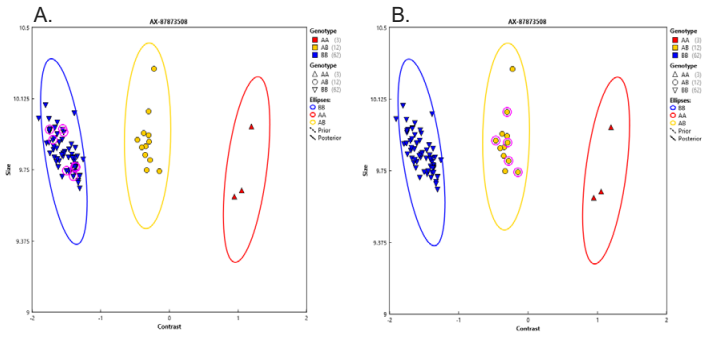


Figure 3. Cluster plots of data obtained from gDNA and WCL. Data points that are highlighted with pink circles are from the same fish, including data points derived from purified gDNA and whole cell lysates. A. Highlighted data points are from samples obtained from Fresh6 (four gDNA and four WCL). B. Highlighted data points are from samples from fish Froze3 (four gDNA and one WCL).

Titration of optimal lysate volumes

The results described above demonstrate that genotyping data obtained from whole cell lysates are very similar to data obtained using purified DNA. To determine what volume of lysate can be used in a SwiftArray analysis, we titrated differing amounts of whole cell lysate into the target preparation workflow. Eleven samples were analyzed in duplicate using 5µl, 10µl, 20µl and 50µl of leftover lysate. Samples with less than 20µl were brought to 20µl total volume using water before initiating the SwiftArray assay workflow. The lysates were processed according to the SwiftArray assay user guide and analyzed on SwiftArrayStudio with 16 hour hybridization as described above.

Interestingly, the amount of input lysate had little effect on any of the QC metrics. With the exception of the Froze3 sample, the amount of DNA recovered after template preparation, the dish QC metrics, and the QC call rate, were not significantly different (Figure 4). Moreover, the percentage of no calls was not significantly different from purified genomic DNA, or from each other (Figure 5). These results suggest that the analysis of whole cell lysates with SwiftArrayStudio may generate comparable data to those achieved when using purified genomic DNA.

A.

	Fresh1	Fresh2	Fresh3	Fresh4	Froze1	Froze2	Froze3	Froze4	Fresh5	Fresh6	Fresh7
5µl	1002	983.8	1143	1049	1086	1075	365.0	1046	1094	1122	983.4
	1302	1043	1105	1070	1057	1035	395.9	1069	1067	1134	1136
10µl	1146	1056	1086	1078	1010	1014	373.1	1098	1035	996.7	1120
	1191	1071	1103	1063	1030	1042	369.5	1022	1060	1148	1096
20µl	1096	1001	1061	1066	1039	1024	387.6	1073	1025	1063	1099
	1042	967.0	1041	1099	994.1	1012	399.0	1120	1033	1092	1120
50µl	1091	907.0	1087	1050	991.9	1005	430.2	1063	1002	1051	1058
	1119	844.2	1052	1109	1077	1062	524.9	1096	1016	1054	1057

B.

	Fresh1	Fresh2	Fresh3	Fresh4	Froze1	Froze2	Froze3	Froze4	Fresh5	Fresh6	Fresh7
5µl	A 0.98485	0.98485	0.98617	0.98485	0.98748	0.98946	0.7556	0.99144	0.98748	0.98551	0.98814
	B 0.98682	0.9809	0.9888	0.97892	0.98748	0.98617	0.84256	0.98814	0.98814	0.98551	0.98485
10µl	C 0.98617	0.98419	0.98617	0.98155	0.98946	0.98485	0.87154	0.98682	0.98814	0.98617	0.98946
	D 0.98617	0.98353	0.99078	0.98287	0.98617	0.98617	0.89065	0.99012	0.98617	0.98748	0.98946
20µl	E 0.98419	0.98617	0.98814	0.98221	0.98814	0.98551	0.91107	0.98682	0.9888	0.98353	0.98946
	F 0.98353	0.9809	0.98748	0.98419	0.98682	0.98748	0.91173	0.99078	0.98814	0.98485	0.98617
50µl	G 0.97958	0.98682	0.98682	0.98221	0.98682	0.98682	0.9473	0.9888	0.98617	0.98617	0.98617
	H 0.9809	0.98682	0.99012	0.98485	0.98617	0.98617	0.96311	0.98748	0.98814	0.98748	0.98419
	1	2	3	4	5	6	7	8	9	10	11

C.

5µl	A 98.895	98.428	99.109	98.727	98.727	98.916		98.785	99.199	99.219	99.277
	B 98.994	98.559	99.078	98.408	98.612	98.727	92.373	98.879	99.225	99.115	99.193
10µl	C 99.068	98.869	99.115	98.785	98.842	98.931	94.358	98.853	99.204	99.073	99.235
	D 99.047	98.874	99.005	98.502	98.727	98.764	94.254	98.853	99.199	99.151	99.34
20µl	E 99.083	98.916	98.643	98.91	98.816	98.727	94.054	98.711	99.251	98.298	99.261
	F 99.057	98.601	99.199	98.743	98.821	98.832	94.295	98.753	99.235	99.125	99.162
50µl	G 99.01	98.795	99.225	98.931	98.638	98.628	94.474	98.853	99.136	99.219	99.141
	H 98.79	98.952	99.225	99.031	98.69	98.895	95.186	98.586	99.225	99.225	99.193
	1	2	3	4	5	6	7	8	9	10	11

Figure 4. QC metrics of varying amounts of whole cell lysate used in SwiftArray assay. Five, 10, 20 or 50µl was used in the assay. A. Amount of DNA recovered (micrograms) after amplification, fragmentation and precipitation in the SwiftArray assay workflow. Note that the amount recovered did not depend on the amount input. Also note that Froze3 sample worked less well than the other samples. B. Dish QC of titrated samples. As expected from the low recovery, Froze3 had lower DQC values. C. QC call rate of titrated samples. Again, the quality of the data did not correlate with the input amount (except for Froze3).

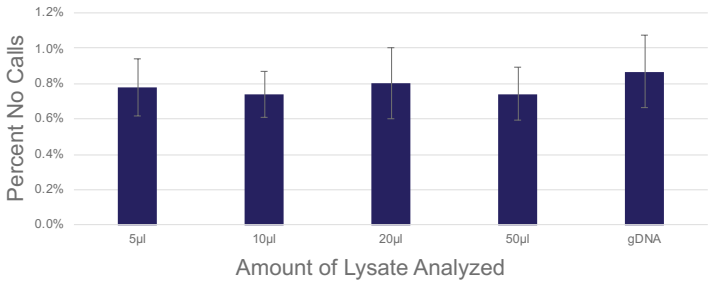


Figure 5. Percent of no calls from whole cell lysates is not significantly different from purified DNA. The percentage no calls was averaged across all the different titration amounts in all the samples, and compared to purified gDNA from the same samples. No significant difference was seen.

Correlation of genotyping data

Finally, we analyzed the congruence of genotypes collected from purified DNA and from whole cell lysates. For each of the probesets on the array, we compared the calls made in purified gDNA to calls made in lysates (Table 3). Overall, average percent of identical calls was 97.49% ± 1.29. If no calls were excluded, the average rose to 99.49% ± 0.56. These data further illustrate that analysis of lysates can produce high-quality data comparable to purified genomic DNA.

The high quality of the data allowed us to establish the identity of the different fish samples. We performed a pairwise correlation of the set of genotypes across all loci in all samples to find which ones were most similar (Figure 6). These data suggest that samples Fresh1 and Fresh3 were from the same fish,

and samples Fresh2 and Fresh4 were from the same fish, but different from Fresh1/3. Samples Fresh5-Fresh10 were all from the same fish. Finally, each of frozen samples appeared to be from different from each other.

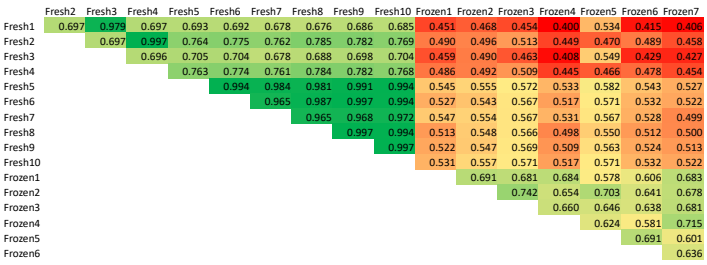


Figure 6. Correlations of genotypes indicate which separately-sold pieces were from the same fish. Fresh1 and Fresh3 were likely identical, Fresh2 and 4 were likely identical. All of Fresh5-10 pieces appear to be from the same fish, whereas each of samples Frozen1-7 appear to be from different fish.

Table 3. Genotypes of individual fish were identical whether they were analyzed by gDNA or whole cell lysate (WCL). The total number of samples analyzed and the numbers that were gDNA or WCL is shown. The % Identical column shows the fraction of probeset calls that were identical in all the samples; the % Identical w/o no calls column shows the fraction of probeset calls that were identical if no calls were excluded. A total of 126,554 probesets were compared.

	n=	% Identical	% Identical w/o no calls
Fresh1	3 (0 DNA, 3 WCL)	97.96	99.77
Fresh2	4 (2 DNA, 2 WCL)	97.64	99.77
Fresh3	4 (2 DNA, 2 WCL)	98.40	99.91
Fresh4	3 (2 DNA, 1 WCL)	97.50	99.62
Fresh5	6 (4 DNA, 2 WCL)	98.11	99.77
Fresh6	8 (4 DNA, 4 WCL)	97.77	99.84
Fresh7	4 (2 DNA, 2 WCL)	96.27	98.61
Fresh8	4 (2 DNA, 2 WCL)	99.02	99.78
Fresh9	4 (2 DNA, 2 WCL)	98.99	99.78
Fresh10	4 (2 DNA, 2 WCL)	98.91	99.73
Froze1	6 (4 DNA, 2 WCL)	95.97	99.14
Froze2	7 (4 DNA, 3 WCL)	93.97	97.98
Froze3	5 (4 DNA, 1 WCL)	97.29	99.75
Froze4	3 (2 DNA, 1 WCL)	97.89	99.74
Froze5	3 (2 DNA, 1 WCL)	96.15	98.63
Froze6	4 (2 DNA, 2 WCL)	97.98	99.83
Froze7	4 (2 DNA, 2 WCL)	97.48	99.75

Summary

In this application note, we demonstrate how the SwiftArraySolution for microarray analysis could be used for genotyping in aquaculture. We showed that the KingFisher Sample Purification system and MagMax CORE Nucleic Acid Purification kit can be used for automated DNA extraction from salmon samples. For those wanting a more hands-off approach, we demonstrated a whole cell lysate method for collecting and analyzing salmon samples. We showed how the Axiom Salmon Genotyping Array for Atlantic salmon can be used with the time-saving SwiftArrayStudio Microarray Analyzer. Finally, we showed that data collected with whole cell lysates was very similar to data collected from purified genomic DNA. Together, these solutions provide aquaculture breeders and stewards with genotyping tools that can help with healthy fish production.

References

1. Houston RD and Macqueen DJ. (2019) Atlantic salmon (*Salmo salar* L.) genetics in the 21st century: taking leaps forward in aquaculture and biological understanding. Anim Genet. 2019 Feb;50(1):3-14. doi: 10.1111/age.12748.
2. Hagen IJ et al. (2026) Genetic markers for tracing introgression of farmed Atlantic salmon (*Salmo salar)* in wild conspecifics. Molecular Ecology Resources 26:e70065 doi: 10.1111/1755-0998.70065
3. Diserud OH et al. (2019) Escaped farmed Atlantic salmon in Norwegian rivers during 1989-2013. ICES Journal of Marine Science 76:1140-1150. doi: 10.1093/icesjms/fsy202
4. Sonesson AK et al. (2023) Sustainable management and improvement of genetic resources for aquaculture. Journal of the World Aquaculture Society, 54(2), 364–396. doi: 10.1111/jwas.12968
5. Houston et al. (2014). Development and validation of a high density SNP genotyping array for Atlantic salmon (*Salmo salar*). BMC Genomics 15:90. doi: 10.1186/1471-2164-15-90
6. Yáñez JM et al. (2016). Genomewide single nucleotide polymorphism discovery in Atlantic salmon (*Salmo salar*): validation in wild and farmed American and European populations. Molecular Ecology Resources 16:1002-1011. doi: 10.1111/1755-0998.12503
7. Barria A et al. (2018) Population genomic structure and genome-wide linkage disequilibrium in farmed Atlantic salmon (*Salmo salar* L.) using dense SNP genotypes. Front. Genet. 9:649. doi: 10.3389/fgene.2018.00649
8. Yáñez JM et al. (2019). Comparative genomic analysis of three salmonid species identifies functional candidate genes involved in resistance to the intracellular bacterium *Piscirickettsia salmonis*. Front. Genet. 10:665. doi: 10.3389/fgene.2019.00665
9. Moghadam HK et al. (2026). From challenge tests to field survival: cross generational genomic selection improves cardiomyopathy syndrome resistance in Atlantic salmon (*Salmo salar*). Aquaculture 622:744087. doi: 10.1016/j.aquaculture.2026.744087
10. Sae-Lim P et al. (2025). Genomic prediction accuracy of growth in Atlantic salmon (*Salmo salar*) when genotype-by-environment interaction is present. Aquaculture 603:742391. doi: 10.1016/j.aquaculture.2025.742391
11. Tollervey MJ et al. (2026). Impact of freshwater rearing on saltwater performance: a genotype-environment interaction study in Atlantic salmon (*Salmo salar*). Aquaculture 610:742892. doi: 10.1016/j.aquaculture.2025.742892

Ordering information

Description	Cat. No
SwiftArray Reagents and Systems	
Applied Biosystems™ Axiom™ Salmon Genotyping Array, 96F	550540
Applied Biosystems™ SwiftArrayStudio™ Microarray Analyzer	00-0471
Applied Biosystems™ Axiom™ SwiftArrayStudio™ Consumables Kit	952674
KingFisher Reagents and Systems	
Thermo Scientific™ KingFisher™ Flex Purification System with 96 Deep-well Head	5400630
Applied Biosystems™ MagMAX™ CORE Nucleic Acid Purification Kit	A32702
Crude Lysate Preparation Reagents	
Applied Biosystems™ AgriSeq™ Genomic DNA Extraction Kit	A66428
Fisherbrand™ RNase-Free Disposable Pellet Pestles	12-141-364

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