Medicinal Genomics SenSATIVAx® and PathoSEEK® on the MGISP-960 High-throughput Automated Sample Preparation System

Automated DNA Isolation and qPCR Setup from Cannabis Matrices

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Introduction

MGISP-960 High-throughput Automated Sample Preparation System is a liquid handling workstation with integrated 96 channel pipette, which can be used for nucleic acid extraction and subsequent quantitative PCR (qPCR) setup. Medicinal Genomics SenSATIVAx Plant/Microbial DNA Purification Kit and PathoSEEK Microbial Safety Testing Solution workflow employs a magnetic bead-based DNA isolation and qPCR technology to test cannabis flower and cannabis infused products for microbial contamination. These kits were automated on the MGISP-960 System.

Automated Method Workflow and Benefits

The automated Medicinal Genomics SenSATIVAx and PathoSEEK workflow features an intuitive user interface that allows for selection of customizable workflow options. The user may process up to 192 samples when running SenSATIVAx and can set up a full 96 well plate of qPCR reactions (including positive and negative controls) when running PathoSEEK. Options selected through the user interface will automatically update and create a deck setup display which guides the user as to where reagents and consumables should be placed.

A full 96-sample extraction can be processed in approximately 60 minutes, and a full 96-reaction qPCR plate can be set up in approximately 20 minutes.

Figure 1. User interface for Medicinal Genomics PathoSEEK qPCR setup method automated on the MGISP - 960 automated sample preparation system.

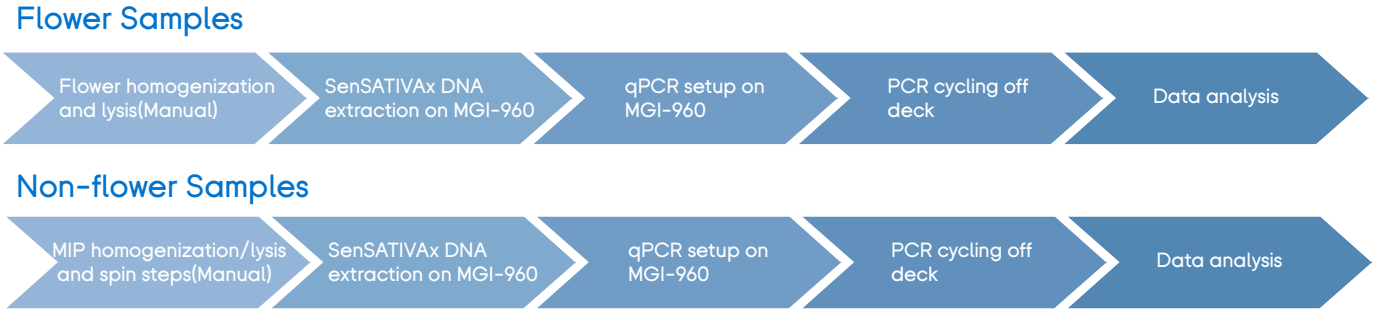

Figure 2. SenSATIVAx and PathoSEEK assay workflows:

Experimental Method

Flower Testing

As defined in the Medicinal Genomics SenSATIVAx and Pathoseek User Guides, 1 g of Cannabis flower sample was weighed into a Whirl-Pak bag (Nasco #B02385WA) followed by the addition of 9.0 mL of tryptic soy broth (TSB, MGC #420205) to the bag. The flower samples were then manually homogenized for 1 minute. Following homogenization, 1 mL of the homogenized flower/TSB mixtures were transferred into a 1.5 mL snap cap tubes. This process was repeated for each of the 6 flower samples being tested. The DNA extracts using SenSATIVAx reagents were processed on the MGISP-960 platform. The same six flower samples were also processed manually.

Samples were then tested for Aspergillus and total yeast and mold contamination via qPCR using the MGC PathoSEEK Aspergillus 2-color Detection assay (MGC #420130), and Total Yeast and Mold Detection Assay (MGC #420103) on the MGISP-960 platform. The qPCR was run on the Agilent AriaMx qPCR instrument. See Table 1 for results.

MIP Testing

AAs defined in the Medicinal Genomics SenSATIVAx for MIP SOP, 1 g each of 2 MIP samples were weighed into 2 separate 15 mL conical tubes followed by the addition of 7 mL of MIP Solution A. The MIP samples were then homogenized by vertexing until in solution. Following homogenization, 1 mL of the homogenized edible samples with MIP solution A were transferred into 1.5 mL snap cap tubes. The internal SCCG control was added to the sample, followed by vortexing to mix and centrifuging. A portion of the samples was transferred into fresh 1.5 mL snap cap tubes followed by the addition of an equal volume of chloroform. The samples were then vortexed vigorously then centrifuged again and a portion of the supernatant was transferred to a 96-well extraction plate. Solution B was added to the sample. This process was performed in parallel for each of the MIP samples being tested. Using SenSATIVAx reagents, the remainder of the DNA extraction was processed on the MGISP-960 platform. The same MIP samples were also processed manually.

Samples were then tested for Aspergillus and total yeast and mold contamination via qPCR using the MGC PathoSEEK 2-Color Aspergillus Multiplex Detection Assay (MGC #420130) and Total Yeast and Mold Detection Assay (MGC #420103) on the MGISP-960 platform. The qPCR was run on the AriaMx qPCR instrument. See Table 1 for results.

Table 1. Automated vs Manual Processing of flower and MIP samples

Matrix	Target	Cq Value(FAM)		Target	Cq Value(HEX)	
		MGI 960	Manual		MGI 960	Manual
Flower	Aspergillus	34.28	35.62	Cannabis DNA	21.72	22.61
Flower	Aspergillus	36.89	No Cq	Cannabis DNA	21.75	22.45
Flower	Aspergillus	No Cq	No Cq	Cannabis DNA	25.06	25.72
Flower	Aspergillus	No Cq	No Cq	Cannabis DNA	24.96	25.65
Flower	Aspergillus	No Cq	No Cq	Cannabis DNA	23.42	24.02
Flower	Aspergillus	No Cq	No Cq	Cannabis DNA	23.62	23.91
Edible	Aspergillus	No Cq	No Cq	Cannabis DNA	30.15	30.19
Edible	Aspergillus	No Cq	No Cq	Cannabis DNA	29.02	28.85
Flower	Total Y&M	No Cq	No Cq	Cannabis DNA	21.58	22.47
Flower	Total Y&M	No Cq	No Cq	Cannabis DNA	21.6	22.1
Flower	Total Y&M	No Cq	No Cq	Cannabis DNA	24.83	25.94
Flower	Total Y&M	No Cq	No Cq	Cannabis DNA	24.76	25.56
Flower	Total Y&M	No Cq	No Cq	Cannabis DNA	23.37	24.09
Flower	Total Y&M	No Cq	No Cq	Cannabis DNA	23.3	23.84
Edible	Total Y&M	No Cq	No Cq	Cannabis DNA	31.83	31.24
Edible	Total Y&M	No Cq	No Cq	Cannabis DNA	28.79	29.87
Asp 2 Color POS Con	Aspergillus	23.3	23.16	Cannabis DNA	No Cq	No Cq
TYM POS Con	Aspergillus	10.15	10.07	Cannabis DNA	No Cq	No Cq
Asp 2 Color NTC	Total Y&M	No Cq	No Cq	Cannabis DNA	No Cq	No Cq
TYM NTC	Total Y&M	No Cq	No Cq	Cannabis DNA	No Cq	No Cq

Conclusion

The Medicinal Genomics SenSATIVAx Plant/Microbial DNA Purification Kit and PathoSEEK Microbial Safety Testing Solution kits automated on the MGISP-960 automated liquid handling platform were demonstrated to provide a robust, flexible, and efficient DNA extraction and qPCR setup workflow for multiple matrices. 96 samples can be extracted in 1 hour and a full 96-well plate of qPCR reactions can be set up in approximately 20 minutes. The intuitive user interface guides the user through the setup process, which allows for seamless setup and method execution.

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