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NYS DOH LEB-603

Preparation of Samples for Medical Marijuana Testing

Table of Contents

1.0. Scope and Application

1.1. This method, NYS DOH LEB-603, Preparation of Samples for Medical Marijuana Testing (ELAP Method ID 9926) describes the preparation of samples of medical marijuana products as required in Title 10 (Health), Subpart 55-2.15 and Chapter XIII, Section 1004.14 of the official Compilation of Codes, Rules, and Regulations, of the State of New York for the detection of microbial contaminants, including bile tolerant gram negative bacteria, *Escherichia coli*, *Clostridium* species, *Pseudomonas* species, *Streptococcus* species, *Salmonella* species, *Penicillium* species, *Aspergillus* species *Mucor* species, and thermophilic actinomycetes. Numbers of fungi and aerobic bacteria are also determined.

1.2. Protocols for the identification of these organisms in samples of medical marijuana products can be found in the NYS DOH LEB-600 series. See Appendix A for Medical Marijuana Microbial Testing Plan flowcharts.

2.0. Summary of the Method

2.1. Twenty doses of individual lots of medical marijuana product are batched: 4 doses are stored for later use, and the remaining 16 doses are diluted 1:10 in PBST and distributed equally into 16 sterile containers. Samples containing materials that are not soluble in water are diluted with isopropyl myristate prior to adding PBST. Matrix spikes of microbial contaminants and positive and negative controls are prepared and analyzed in parallel with sample aliquots.

3.0. Definitions

3.1. PBST stands for Phosphate Buffered Saline, pH 7.2, containing 0.1% Tween® 80.

3.2. Polysorbate 80 (Tween® 80) is a nonionic surfactant and emulsifier.

3.3. Batched product refers to the combining of individual sample aliquots from a single lot of product into one sample to be tested.

3.4. BioBalls® are small water-soluble balls containing a known number of microorganisms that are used for quality control.

3.5. SM refers to *Standard Methods for the Analysis of Water and Wastewater*.

3.6. BSC stands for biological safety cabinet.

4.0. Health and Safety Warnings

4.1. Microbiological analyses involve the culturing of potentially pathogenic organisms.

4.1.1. All microbiologically contaminated media in the laboratory shall be autoclaved prior to disposal.

4.1.2. Laboratory equipment and benches shall be disinfected before and after use with a minimum concentration of 75% ethanol.

4.1.3. Mouth pipetting is prohibited.

4.1.4. All accidents, particularly those which may result in infection, shall be reported according to laboratory specific policies and procedures.

4.2. Laboratory safety procedures shall be followed at all times. Regulations required by federal, state and local government agencies shall be implemented and followed.

4.3. The analyst must be familiar with any possible hazards from the reagents and standards used for sample preparation and analysis.

4.4. Always follow guidelines listed in safety data sheets (SDS) for proper storage, handling, and disposal of samples, solvents, reagents, and standards. SDS are located within the laboratory. These guidelines must be made available to all personnel involved in microbiological analyses.

4.5. Appropriate lab coat, safety glasses and gloves must be worn when performing standard or sample preparations, working on instrumentation, disposing of waste, and cleaning laboratory equipment.

5.0. Handling and Preservation

5.1. Sample shipping conditions - The medical marijuana products from the Registered Organizations (ROs) are shipped as per manufacturer's specifications and must adhere to all regulatory requirements.

5.2. Sample Receipt - Medical marijuana products from the RO are received, verified and documented ensuring that method, regulatory and Accreditation Body requirements are met. All medical marijuana products must be stored under the conditions based on the manufacturer's recommendation. The storage is documented.

5.3. Method holding times

5.3.1. Once samples are prepared, they are analyzed immediately following NYS DOH LEB-604, NYS DOH LEB-605 and NYS DOH LEB-609.

5.4. Preservation

5.4.1. Samples diluted in PBST that are not required for analyses are stored refrigerated until it has been determined that they are not needed for additional microbiological evaluation.

5.5. Storage

5.5.1. Samples are analyzed upon receipt.

5.5.2. If storage is required, samples are maintained at room temperature in a secure location.

6.0. Interferences

6.1. The presence of spreading colonies or confluent growth can interfere with accurate colony enumeration.

7.0. Apparatus and Materials

7.1. Equipment and Supplies

7.1.1. Water bath, 30-35°C

7.1.2. Sterile tubes, various sizes

7.1.3. Measuring device for determining volumes, e.g., sterile pipets and graduated cylinders

7.1.4. Automatic pipetters and sterile tips

7.1.5. Pipets, sterile disposable, 5 mL, 10 mL, 25 mL.

7.1.6. Balance

7.1.7. Tweezers and ethanol for flame-sterilization

7.1.8. Bunsen burner

7.1.9. Weights traceable to NIST

7.2. Reagents and Chemicals

7.2.1. Phosphate Buffered Saline pH 7.2, 10 and 100 mL aliquots in tubes/bottles, containing 0.1% Tween 80 (PBST)

7.2.2. Isopropyl myristate, Sigma Chemical Company, cat. no. M0757 or equivalent

7.2.3. *Aspergillus brasiliensis* (also known as *A. niger*) ATCC 16404, BioBall® MultiShot 550, bioMérieux cat. no. 56001 or 56011.

7.2.4. *Clostridium sporogenes* ATCC 11437, BioBall® MultiShot 550, bioMérieux cat. no. 56004 or 56014.

7.2.5. *Enterococcus faecalis* ATCC 29212, either lab prepared spiking solution or BioBall® MultiShot 550, bioMérieux cat. no. 56005 or 56015.

7.2.6. *Escherichia coli* ATCC 8739, either lab prepared spiking solution or BioBall® MultiShot 550, bioMérieux cat. no. 56006 or 56016.

7.2.7. *Klebsiella pneumoniae* ATCC 13833, lab prepared spiking solution.

7.2.8. *Pseudomonas aeruginosa* ATCC 9027, either lab prepared spiking solution or BioBall® MultiShot 550, bioMérieux cat. no. 56007 or 56017.

7.2.9. *Salmonella typhimurium* ATCC 14028, either lab prepared spiking solution or BioBall® SingleShot, bioMérieux cat. no. 413775 or 56044.

7.2.10. *Staphylococcus aureus* ATCC 6538, either lab prepared spiking solution or BioBall® MultiShot 550, bioMérieux cat. no. 56009 or 56019.

7.2.11. SM dilution water, prepared according to 9050C.

7.3. Forms

7.3.1. Sample Preparation Log (e.g. LEB-RS-603A, Appendix B)

8.0. Quality Control/Assurance (Laboratories must conform to sections 9020-9050 of Standard Methods for the Examination of Water and Wastewater.)

8.1. Method Detection Limits

8.1.1. Detection limits are determined in accordance with relevant standards, regulations and accreditation body requirements.

8.2. Calibration and Standardization

8.2.1. Temperatures of the cold room and refrigerators/freezers are observed and recorded at least daily.

8.2.1.1. If the cold room or refrigerator does not stay within 1-8°C, the supervisor is notified.

8.2.1.2. The optimum temperature range for a refrigerator is 1-4°C

8.2.1.3. If the cold room or refrigerator was in a defrost cycle at the time that the temperature was recorded, and the temperature does not reach 8°C, re-testing of media is not required.

8.2.1.4. Media may be re-tested for quality, depending on the number of degrees and the amount of time that the cold room temperature was out of compliance, at the discretion of the laboratory.

8.2.2. Temperature of the freezer is observed and recorded at least daily. If the freezer exceeds -15°C, the laboratory specific corrective actions are followed.

8.2.3. Water bath temperatures shall be observed and recorded twice daily, separated by at least 4 hours.

8.2.3.1. Temperature of the 30-35°C water bath is observed and recorded.

8.2.3.1.1. If the water bath temperature does not stay within 30-35°C, laboratory specific correction actions are followed. Analytical results may be invalidated if the water bath temperature exceeds 40.0°C.

8.2.4. Max/min temperatures are recorded when daily temperature measurements are not possible, such as on holidays and weekends.

8.2.4.1. Max/min temperatures of freezers may show transient excursions of temperature above 0°C due to defrost cycles.

8.2.4.2. This is acceptable, provided that the quality of BioBalls® are unaffected.

8.2.5. Thermometers must be calibrated as prescribed by the Accreditation Body and in accordance with relevant regulations and standards.

8.2.6. Sterility of disposable loops and spreaders is determined as prescribed by the Accreditation Body and in accordance with relevant regulations and standards.

8.2.7. Micropipetters are calibrated as prescribed by the Accreditation Body and in accordance with relevant regulations and standards.

8.3. Quality Control

8.3.1. Invalidate lot of media if tests are not in accordance with acceptance criteria as prescribed by the Accreditation Body and in accordance with relevant regulations and standards.

8.3.2. Acceptability of PBST is determined as prescribed by the Accreditation Body and in accordance with relevant regulations and standards.

8.3.3. Acceptability of supplies is tested according to requirements prescribed by the laboratory, Accreditation Body and in accordance with relevant regulations and standards.

8.3.4. The use test for deionized water is performed annually, when cartridges are changed, or repairs are made to the deionized water systems and as prescribed by the laboratory, Accreditation Body and in accordance with relevant regulations and standards.

8.3.5. Liquid media shall be stored in tightly-capped bottles in the dark at 4°C for up to 3 months from the date of preparation.

8.3.6. Agar plates can be used for 2 weeks if stored refrigerated in plastic bags and in the dark.

8.3.6.1. Agar plates can be used after 2 weeks storage if ongoing QC demonstrates no loss in selectivity or growth promotion.

8.4. Corrective/Preventive Actions

8.4.1. The laboratory will initiate non-conformances and/or corrective/preventive actions in accordance with laboratory specific procedures and as prescribed by the Accreditation Body and in accordance with relevant regulations and standards.

9.0. Procedure

9.1. General

9.1.1. Aseptic technique is used for all procedures.

9.1.1.1. Aseptic technique can be found in a general microbiology textbook or on-line.

9.1.2. On the Medical Marijuana Sample Preparation Log (e.g. LEB-RS-603A) record the sample accession number, sample type analyst initials and date.

9.1.2.1. Twenty dosage units should be removed.

9.1.2.1.1. If 20 doses are not available, record the number of doses removed.

9.1.2.1.2. Ideally use at least 10 mL or 10 g of the product to be examined.

9.1.2.1.3. Sample aliquots from the same production date/lot number are batched prior to analysis.

9.1.2.2. Twenty tubes or flasks are required.

9.1.2.2.1. Four tubes are controls, and do not contain sample.

9.1.2.2.2. Sixteen tubes or flasks will contain sample. Of these, 7 tubes are used as matrix spikes and inoculated with either rehydrated BioBalls® or suspensions of lab prepared organisms (see LEB-AP-603A, Appendix C)

9.2. Preparation of Spiking Solutions.

9.2.1. Prior to any sample preparation or analysis, the spiking solutions must be prepared either in the laboratory or by using BioBalls®.

9.2.2. Laboratory Prepared Spiking Solutions

9.2.2.1. Lab-prepared spikes are diluted from working stock cultures according to standard microbiological procedures.

9.2.2.2. Dilutions are prepared in SM dilution water.

9.2.3. BioBall® Rehydration

9.2.3.1. Remove PBST from the refrigerator and allow to warm to room temperature.

9.2.3.2. For each BioBall® label a 1.7 mL sterile microcentrifuge tube.

9.2.3.3. Add the appropriate volume of PBST to each tube (see table below).

9.2.3.4. Remove the BioBall® vial from the freezer, slowly uncap, and tip the BioBall into the tube containing PBST.

9.2.3.5. Once the BioBall® dissolves, invert the tube several times to mix gently.

9.2.3.6. Incubate the tubes for 4 hours at room temperature.

9.2.3.7. Vortex to mix prior to use.

9.2.3.8. The reconstituted BioBall® can now be used in 100 µL aliquots for media testing.

9.2.3.9. BioBall® preparations of *K. pneumoniae* and *T. vulgaris* are not available.

Organism	Diluent	Spike	Final concentration
Asp. brasiliensis	1.1 mL PBST	MultiShot BioBalls®	50 organisms/0.1 mL
Cl. sporogenes	1.1 mL PBST	MultiShot BioBalls®	50 organisms/0.1 mL
Ent. faecalis	1.1 mL PBST	MultiShot BioBalls®	50 organisms/0.1 mL
E. coli	1.1 mL PBST	MultiShot BioBalls®	50 organisms/0.1 mL
Ps. aeruginosa	1.1 mL PBST	MultiShot BioBalls®	50 organisms/0.1 mL
S. typhimurium	0.1 mL PBST	SingleShot BioBalls®	30 organisms/0.1 mL

9.3. Preparation of Products Soluble in Water

9.3.1. General.

9.3.1.1. In a BSC, aseptically add 20 sample aliquots from the same production date/lot number to a sterile vessel that has been tared.

9.3.1.2. Remove an equal number of doses from each container of sample submitted.

9.3.1.2.1. For example, if 5 bottles of a lot are submitted, remove and batch 4 doses from each bottle.

9.3.1.3. Determine and record the combined sample weight (and if applicable, volume) of the batched aliquots (e.g. LEB-RS-603A).

9.3.1.4. Mix the batched sample well.

9.3.2. Capsules/pills.

9.3.2.1. Remove 20% of the batched sample (4 doses) for archiving.

9.3.2.2. Calculate the volume of PBST required to produce a ten-fold dilution of the remaining 16 doses (weight/volume for pills or solids).

9.3.2.3. Add the calculated volume of PBST to the batched sample and mix well.

9.3.2.4. Measure and record the final sample volume (e.g. LEB-RS-603A).

9.3.2.4.1. Pills or gelcaps may require heating to enhance dissolution.

9.3.2.4.1.1. If required, heat sample to 30-35°C in a water bath with intermittent mixing until the capsules are dissolved.

9.3.2.5. Divide the diluted sample volume by 16 and aliquot equally into 16 sterile tubes or flasks.

9.3.2.6. Record the aliquot volume (e.g. LEB-RS-603A).

9.3.3. Syrups.

9.3.3.1. Remove 20% of the batched sample (4 doses) for archiving.

9.3.3.2. Aliquot one dose of sample into each of 16 flasks.

9.3.3.3. Calculate the volume of PBST required to produce a ten-fold dilution for one dose.

9.3.3.3.1. Take into account that a volume of 10X TSB that is equal to the sample volume will be added during analysis.

9.3.3.3.1.1. For example, for a 15 mL dose, add 120 mLs PBST. Fifteen mLs of TSB will be added subsequently (see NYS DOH LEB-604).

9.3.3.4. Add a volume of PBST to each flask to produce a ten-fold dilution.

9.3.3.5. Record the aliquot volume (e.g. LEB-RS-603A).

9.3.4. Tinctures and oral sprays.

9.3.4.1. Remove 20% of the batched sample (4 doses) for archiving.

9.3.4.2. Calculate the volume of PBST required to produce a ten-fold dilution of the remaining 16 doses (volume/volume).

9.3.4.3. Add the calculated volume of PBST to the batched sample and mix well.

9.3.4.4. Measure and record the final sample volume (e.g. LEB-RS-603A).

9.3.5. Vaporizer pens.

9.3.5.1. Calculate the volume of PBST required to produce a ten-fold dilution of the product (weight/volume).

9.3.5.2. Add the calculated volume of PBST to the batched sample and mix well.

9.3.5.3. Distribute 16 equal aliquots into tubes.

9.3.5.4. Refrigerate remaining diluted product.

9.3.5.5. Measure and record the final sample volume (e.g. LEB-RS-603A).

9.4. Preparation Products Insoluble in Water.

9.4.1. Aseptically add all sample aliquots from the same production date/lot number to a sterile vessel that has been tared.

9.4.2. Determine and record the combined sample weight (and if applicable, volume) of the batched aliquots (e.g. LEB-RS-603A).

9.4.3. If an emulsion cannot be created when the batched sample is diluted with PBST, add an equal volume of filter-sterilized isopropyl myristate (pre-warmed to 30-35°C) to the batched sample prior to dilution.

9.4.4. For example, product contained in vaporizer pens may require the addition of isopropyl myristate in order to create an emulsion.

9.4.5. Warm the batched sample/isopropyl myristate mixture to 30-35°C in a heat block or water bath.

9.4.5.1. Mix carefully.

9.4.5.2. Do not heat above 35°C.

9.4.6. Calculate the volume of PBST (pre-warmed to 30-35°C) required to produce a five-fold dilution of the sample/isopropyl myristate mixture in each aliquot.

9.4.7. For example, if the sample volume is 1 mL and the isopropyl myristate volume is 1 mL, add 8 mLs of PBST.

9.4.8. Add the calculated volume of pre-warmed PBST to each aliquot and vortex. Keep the diluted aliquots warm at 30-35°C until analysis.

9.4.9. Record the final aliquot volume (e.g. LEB-RS-603A).

9.5. Preparation of Positive and Negative Controls.

9.5.1. Prepare a positive control sample for aerobic plate counts by adding a 0.1mL aliquot of *E. coli* BioBalls® or laboratory prepared spiking solution to a tube containing one mL PBST.

9.5.1.1. The PBST aliquot being plated should contain less than 100 CFU *E. coli*.

9.5.2. Prepare a positive control sample for mold plate counts by adding a 0.1mL aliquot of *A. brasieliensis* BioBalls® to a tube containing 1 mL PBST

9.5.2.1. The PBST aliquot being plated should contain less than 100 CFU *A. brasiliensis*.

9.5.3. Prepare two negative control samples by adding 1 mL PBST to two tubes.

9.5.3.1. Neither spiking solutions nor sample are added to negative controls.

9.6. Analysis of product.

9.6.1. Once the samples and controls have been batched, aliquoted, and diluted, and the BioBalls® have rehydrated for 4 hours, proceed with NYS DOH LEB-604, NYS DOH LEB-605 and NYS LEB-609.

9.6.1.1. Sample analyses required are summarized in LEB-AP-603A, Appendix C.

10.0. Data Acquisition, Reduction, Analysis, Calculations, Acceptance Criteria and Documentation

10.1. Record the sample accession number, sample type, total amount of sample received in grams or mLs, final sample volume in PBST or isopropyl myristate/PBST,

expiration dates of PBST and isopropyl myristate, analyst initials and date (e.g. LEB-RS-603A).

11.0. Method Performance

11.1. Demonstration of Capability

11.1.1. Prior to acceptance and use of this method for data reporting, a satisfactory initial demonstration of capability (DOC) is required. Thereafter, an ongoing DOC is to be performed annually.

11.1.2. An initial DOC shall be made prior to using any method, and at any time there is a change in instrument type, personnel or method or any time that a method has not been performed by the laboratory or analyst in a twelve (12) month period.

11.1.3. All DOCs shall be documented, and all data applicable to the demonstration shall be retained and readily available at the laboratory. Consult state regulations and standards for additional information on performing a DOC for microbiological contaminants.

11.1.4. Consult relevant standards, regulations and Accreditation Body requirements for additional information on performing DOCs for microbial contaminants.

11.2. Laboratory Detection Limits

11.2.1. Detection limits are determined in accordance with relevant standards, regulations and accreditation body requirements.

12.0. Waste Management/Pollution Prevention

12.1. It is the responsibility of the laboratory to comply with all federal, state and local regulations governing waste management, particularly the hazardous waste identification rules and land disposal restrictions.

12.2. Bacterial/fungal cultures and contaminated or potentially contaminated disposable materials are disposed of in biohazardous waste cans and autoclaved prior to discarding.

12.3. Dispose of non-hazardous water waste in the laboratory sink followed by flushing with tap water.

12.4. Dispose of glassware in appropriately labeled boxes. Be sure that, whenever possible, the glass has been thoroughly rinsed and is contaminant-free before disposal.

12.5. Consult federal, state and local regulations for additional information or for information on the disposal of products not described in this method.

13.0. References

13.1. United States Pharmacopeia. USP38-NF33, The United States Pharmacopeial Convention, General chapters <61>, <62>, <1111>.

13.2. Standard Methods for the Examination of Water and Wastewater, sections 9020-9050

13.3. NYS DOH LEB-604, Microbial Presence/Absence Test for Medical Marijuana Samples

13.4. NYS DOH LEB-605, Aerobic Plate Counts for Medical Marijuana Testing

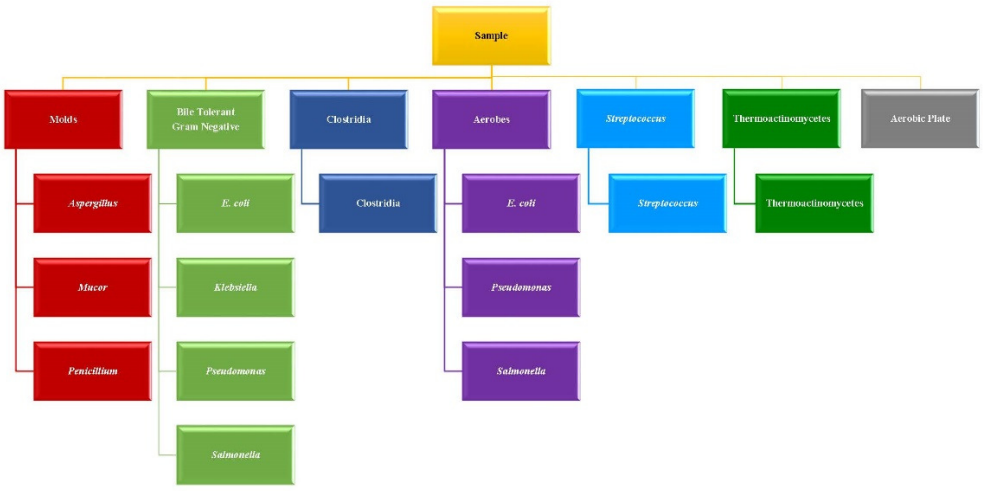
13.5. NYS DOH LEB-609, Mold Plates Counts and Identification for Medical Marijuana Testing

14.0. Appendices

Appendix A – Flowcharts

Medical Marijuana Microbial Testing Plan

Organisms



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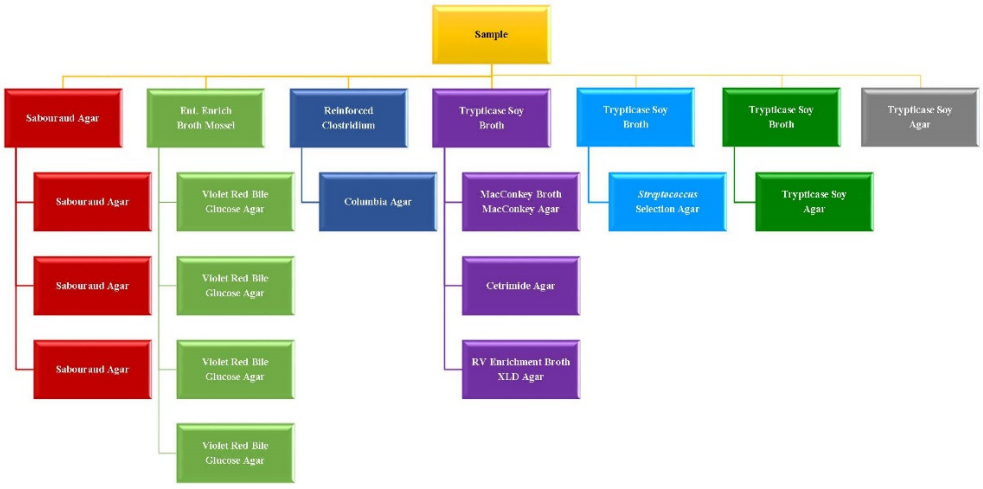
Medical Marijuana Microbial Testing Plan

Media



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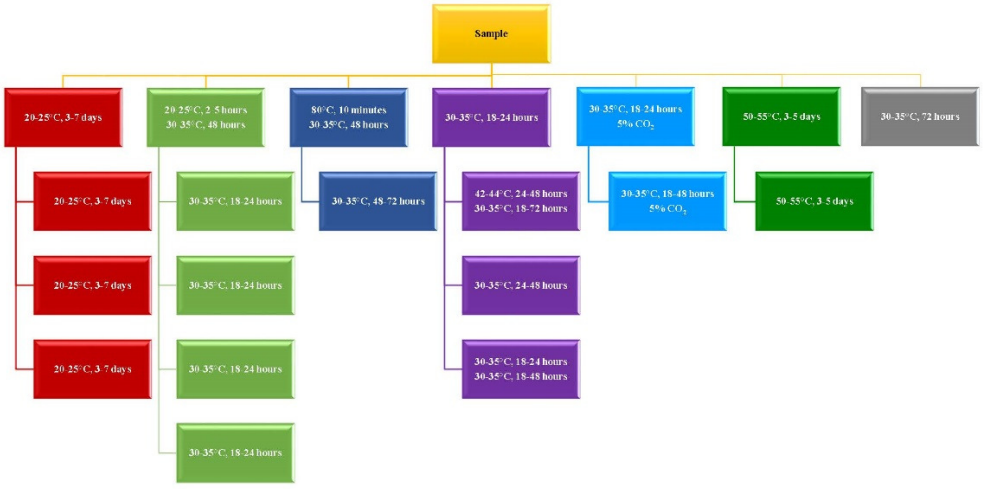
Medical Marijuana Microbial Testing Plan

Incubation Temperatures and Times



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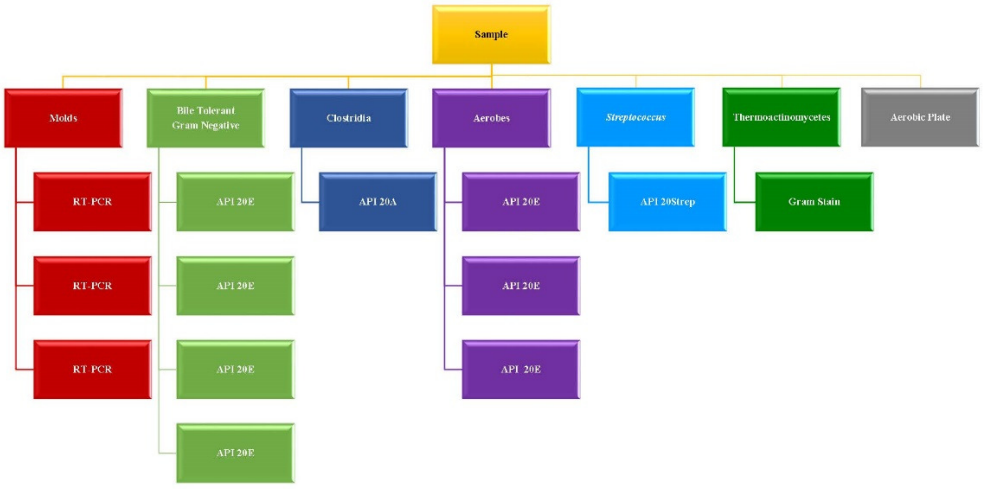
Medical Marijuana Microbial Testing Plan

Colony Identification Assays



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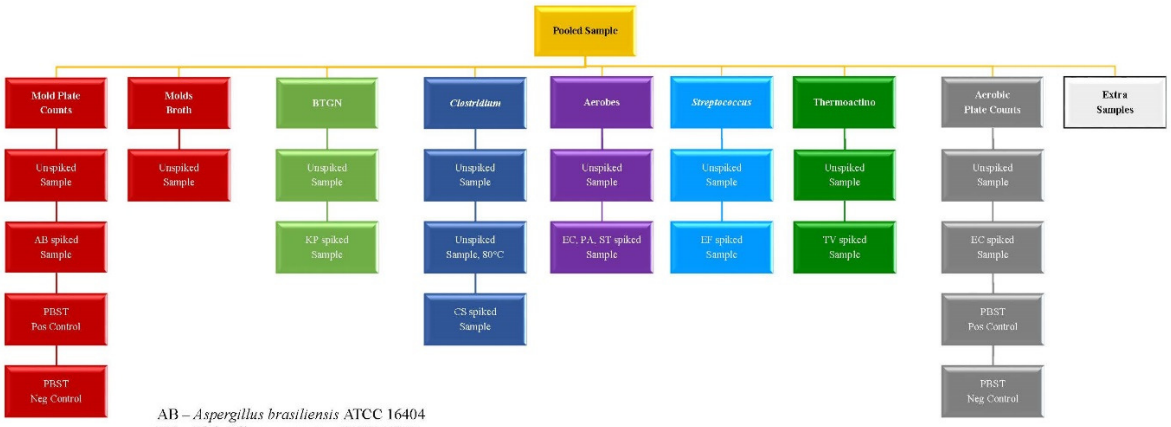
Medical Marijuana Microbial Testing Plan

General Set-Up for Presence/Absence and Plate Count Assays



AB – *Aspergillus brasiliensis* ATCC 16404
KP – *Klebsiella pneumoniae* ATCC 13883
CS – *Clostridium sporogenes* ATCC 11437
EC – *Escherichia coli* ATCC 8739
PA – *Pseudomonas aeruginosa* ATCC 9027
ST – *Salmonella typhimurium* ATCC 14028
EF – *Enterococcus faecalis* ATCC 29212
TV – *Thermoactinomyces vulgaris* ATCC 43649

20 doses pooled
16 doses tested
4 doses stored

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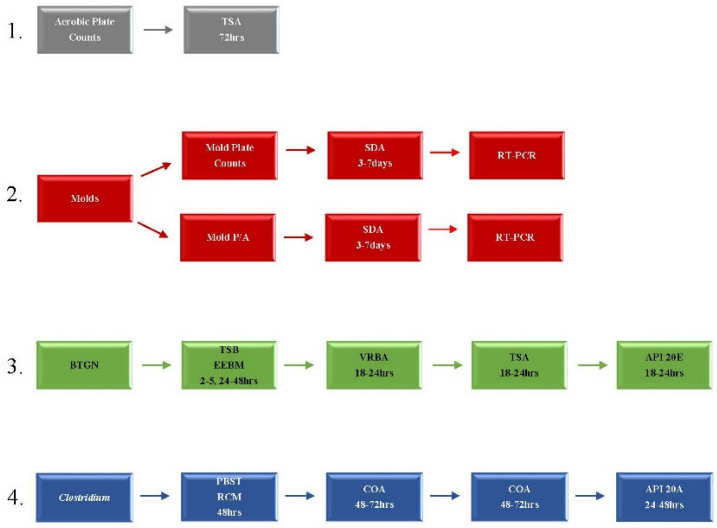
Medical Marijuana Microbial Testing Plan

General Set-Up for Presence/Absence, Colony Identifications, and Plate Count Assays



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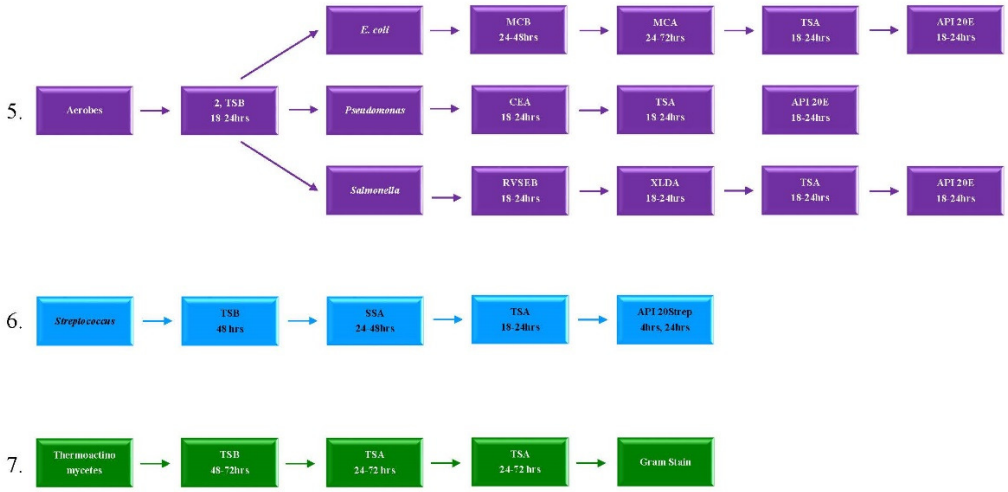
Medical Marijuana Microbial Testing Plan

General Set-Up for Presence/Absence, Colony Identifications, and Plate Count Assays



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Appendix B – Form(s)

Medical Marijuana Sample Preparation Log (LEB-RS-603A)

Accession #	Sample Type	Dose Amount	No. of Batched Aliquots	Combined Aliquot Weight	Diluted Aliquot Volume	PBST Lot Date	Isopropyl Myristate Lot	Analyst Initials/ Date	Comments

_____

Reviewed by Date

Appendix C – Reference (LEB-AP-603A)

Summary of Sample Preparation

Aliquot number	Sample	Target	Matrix spike	Medium	Temperature
1	Yes	Aerobic bacteria	n/a	TSA	30-35 °C
2	Yes	Aerobic bacteria, MS	EC BioBall®	TSA	30-35 °C
3	NO	Aerobic bacteria, pos. control	EC BioBall®	TSA	30-35 °C
4	NO	Aerobic bacteria, neg control	n/a	TSA	30-35 °C
5	Yes	Fungi	n/a	SDA	20-25 °C
6	Yes	Fungi, MS	AB BioBall®	SDA	20-25 °C
7	NO	Fungi, pos. control	AB BioBall®	SDA	20-25 °C
8	NO	Fungi, neg. control	n/a	SDA	20-25 °C
9	Yes	Bile tolerant gram negative organisms	n/a	EEBM	30-35 °C
10	Yes	Bile tolerant gram negative organisms, MS	KP lab suspension	EEBM	30-35 °C
11	Yes	*E coli, Salmonella sp., Pseudomonas sp.*	n/a	TSB	30-35 °C
12	Yes	*E coli, Salmonella sp., Pseudomonas sp.* , MS	EC, ST, PA BioBalls®	TSB	30-35 °C
13	Yes	*Clostridium sp.*	n/a	RCM	30-35 °C, anaerobic
14	Yes	*Clostridium sp.**	n/a	RCM	30-35 °C, anaerobic
15	Yes	*Clostridium sp.*, MS	CS BioBall®	RCM	30-35 °C, anaerobic
16	Yes	*Streptococcus sp.*	n/a	TSB	30-35 °C 5% CO_2
17	Yes	*Streptococcus sp.*, MS	EF BioBall®	TSB	30-35 °C 5% CO_2
18	Yes	Thermophilic actinomyetes	n/a	TSB	50-55 °C
19	Yes	Thermophilic actinomyetes, MS	TV spore suspension	TSB	50-55 °C
20	Yes	Fungi	n/a	SDB	20-25 °C
21	Yes	Archived sample	n/a	n/a	4 °C
22	Yes	Archived sample	n/a	n/a	4 °C
23	Yes	Archived sample	n/a	n/a	4 °C
24	Yes	Archived sample	n/a	n/a	4 °C

AB = *Aspergillus brasiliensis*, CS = *Clostridium sporogenes*, EC = *E. coli*, KP = *Klebsiella pneumoniae*, PA = *Pseudomonas aeruginosa* ST = *Salmonella typhimurium*, and TV=*Thermoactinomyces vulgaris*, MS = matrix spike, TSB = tryptic soy broth, TSA = tryptic soy agar, SDA = Sabouraud Dextrose agar, SDB = Sabouraud Dextrose broth, EEBM = Enterobacterial Enrichment Broth Mossel, RCM = Reinforced Clostridial Medium.

Matrix spikes are prepared according to 9.2, and should contain less than 100 organisms. Refer to organism-specific SOPs for details.

*Heat sample at 80 °C for 10 minutes.