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## Test/QA Plan for Mold-Resistant Gypsum Board Testing

Prepared by:

Research Triangle Institute Research Triangle Park, NC

Approved by:

RTI Project Manager: <u>"Signed by Karin K. Foarde on January 31, 2007"</u> Karin K. Foarde

RTI Quality Assurance Manager: <u>"Signed by W. Cary Eaton on January 31, 2007"</u> W. Cary Eaton

EPA Project Officer:"Signed by Timothy Dean on September 12, 2008"Timothy Dean

EPA Quality Assurance Manager: <u>"Signed by Robert Wright on September 12, 2008"</u> Robert Wright This document serves as the Test and Quality Assurance (QA) plan for mold-resistant gypsum board testing. The Quality Management Plan (QMP) under which this work is conducted is the *Verification Testing of Air Pollution Control Technology Quality Management Plan*, Revision 2.2, February 17, 2005<sup>1</sup>. This QMP was approved by EPA for the ETV program and fulfills the requirements of the ETV program-level QMP.

#### Acknowledgement

RTI would like to thank the companies who provided funding for the test method development: American Gypsum, Georgia-Pacific Gypsum Corporation, Gypsum Association, and National Gypsum. In addition, RTI appreciates the stakeholders and gypsum manufacturers who input to this program.

#### Stakeholders

Tajah Blackburn, U.S. EPA Doris Betancourt, U.S. EPA Elliott Horner, Air Quality Sciences Alison Kinn, U.S. EPA Laura Kolb, U.S. EPA Russ Jerusik, Hercules David Marciniak, General Services Administration Janet Macher, CA Department of Health Services Marc Menetrez, U.S. EPA Tony Worthan, Air Quality Sciences Robert Thompson, U.S. EPA

#### Vendors

Leo Bissonnette, Georgia-Pacific Gypsum Robert Bell, USG Corporation Charles Byers, USG Corporation Kendall Clark, Georgia-Pacific Gypsum John College, BPB America Michael Danks, American Gypsum John Donohue, USG Corporation David Drummond, National Gypsum Jessica Noggle, Georgia-Pacific Gypsum Paul Shipp, USG Corporation Rick Weir, National Gypsum Robert Wessel, Gypsum Association Wayne Wilson, LaFarge

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#### **A3: Distribution List**

<u>EPA</u>

Dr. Timothy Dean Dr. Marc Menetrez Dr. Doris Betancourt Mr. Robert Wright

Research Triangle Institute Ms. Karin Foarde Ms. Deborah Franke Dr. Jonathan Black Dr. Keith Esch Mr. Michael Herman Ms. Lauren Pugh Ms. Tricia Schwartz Dr. W. Cary Eaton Page vi

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ACH	air changes per hour
ADQ	audit of data quality
AQS	Air Quality Sciences
ASTM	American Society for Testing and Materials
a <sub>w</sub>	water activity
CFU	colony forming unit
DNPH	2,4-dinitrophenylhydrazine
DQO	data quality objective
EPA	U.S. Environmental Protection Agency
ESTE	environmental and sustainable technology evaluations
ERH	equilibrium relative humidity
ETV	environmental technology verification
g	gram(s)
GC/MS	gas chromatography/mass spectrometry
ISO	International Organization for Standardization
MC	moisture content
ML	microbiology laboratories
ML SOP	microbiology laboratory standard operating procedure
QA	quality assurance
QAM	quality assurance manager
QAPP	quality assurance project plan
QC	quality control
QMP	quality management plan
RH	relative humidity
RTI	Research Triangle Institute (RTI International)
sec	second(s)
SOP	standard operating procedure
spp	species
t	temperature
ТОР	technical operating procedure
T/QAP	test/quality assurance plan
TSA	technical system audit
TVOC	total volatile organic compounds
VOC	volatile organic compounds
μg	microgram(s)
μm	micrometer(s)

### List of Acronyms/Abbreviations/Definitions

# SECTION A: PROJECT MANAGEMENT

### A4: Project/Task Organization

The Environmental Technology Verification (ETV) Program was established by the Office of Research and Development of the U.S. Environmental Protection Agency to accelerate the development and commercialization of improved environmental technologies through third party verification and reporting of performance. The Environmental and Sustainable Technology Evaluations (ESTE) projects respond directly to EPA's need for credible performance information on technologies that can address high-risk environmental problems. RTI will perform the testing, evaluate the data, and prepare the verification reports. The various quality assurance (QA) and management responsibilities are divided between EPA and RTI key project personnel as defined below. The lines of authority between key personnel for this project are shown on the project organization chart in Figure 1.

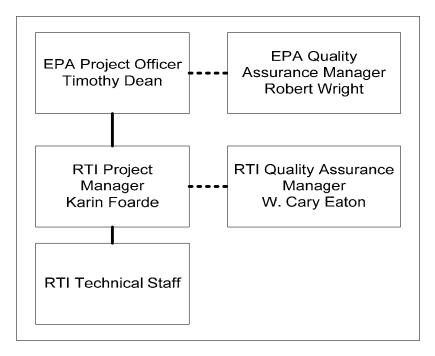


Figure 1. Organization Chart. Dotted lines indicate organizational independence.

## A4.1: Management Responsibilities

Project management responsibilities are divided between EPA and RTI personnel as listed below.

## A4.1.1: EPA Project Officer

Timothy Dean is the EPA Project Officer for the contract. He is responsible for oversight of this program.

# A4.1.2: RTI Project Manager

The RTI Project Manager Karin Foarde is responsible for task implementation and technical quality control. The RTI Project Manager is also responsible for the following:

• Update and deliver revisions of the Test/Quality Assurance Project Plan (T/QAP),

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- Define task objectives in coordination with EPA,
- Develop a detailed work plan schedule,
- Halt testing if there is a quality or safety problem,
- Work with vendors and stakeholders,
- Provide oversight of AQS work,
- Review work progress to ensure that task budgets and schedules are met, and
- Prepare verification reports.

Karin Foarde will also be the technical leader and will review and compare the results for consistency based on her experience with the respective components of the tests.

# A4.2: Quality Assurance Responsibilities

QA responsibilities are divided between the EPA and RTI personnel as listed below.

# A4.2.1: EPA Quality Assurance Manager

The EPA Quality Assurance Manager (EPA QAM) Robert Wright will conduct audits of RTI's QA System<sup>1</sup> and of specific technical activities on the project as specified by EPA. He will be available to resolve any QA issues relating to performance and EPA QA requirements. Specific functions and duties of the EPA QAM include approving the contents of this T/QAP and subsequent revisions, performing a TSA (technical system audit) on RTI, and reviewing QA reports prepared by RTI, including RTI's internal QA evaluations and audits.

# A4.2.3: RTI Quality Assurance Manager

The RTI Quality Assurance Manager (RTI QAM) W. Cary Eaton is organizationally independent of the RTI Project Manager and is responsible for ensuring that QA/quality control (QC) procedures described in this T/QAP are followed. In addition, the RTI QAM will:

- Maintain regular communication with the EPA QAM regarding QA issues,
- Report on the adequacy, status, and effectiveness of the QA program to the Project Manager,
- Conduct an internal TSA after samples have been received and cut for distribution. This will constitute a "readiness review." A conversation will also be held among QAM, AQS contact, and RTI Project Manager to inquire about the AQS quality system. An audit of data quality (ADQ) will be conducted after results from day zero are in hand. Any TSA items not completed during "readiness review" will be completed at the time of the ADQ.
- Provide QA oversight of AQS work,
- Halt testing if a quality or safety problem is found in the TSA, after consultation with RTI program manager,
- Ensure that corrective action, if necessary, is properly implemented and documented,
- Review and approve test (including QC) reports, and
- Prepare the QA section of each verification report.

# A5: Problem Definition/Background Information

Fungal growth and the resulting contamination of building materials is a well-documented problem, especially after the reports from New Orleans and the U.S. Gulf Coast post Hurricane Katrina. However, contaminated materials have been recognized as important indoor fungal reservoirs for years. For example, contamination with fungi has been associated with a variety of materials including carpet, ceiling tile, gypsum board, wallpaper, flooring, insulation, and heating, ventilation and air conditioning components<sup>2,3,4,5</sup>.

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Exposure to fungi may result in respiratory symptoms of both the upper and lower respiratory tract such as allergy and asthma<sup>6</sup>. Everyone is potentially susceptible. However, of particular concern are children with their immature immune systems and individuals of all ages that are immunocompromised<sup>7,8</sup>.

One approach to limiting exposure is to reduce the levels of fungi in the indoor space. For some

sensitive individuals, limiting exposure through avoidance is an effective control method; however, avoidance is not always possible or practical. The investigation, development, and application of effective source controls and strategies are essential to prevent fungal growth in the indoor environment. Mold resistant wallboard is a potentially effective method of source control.

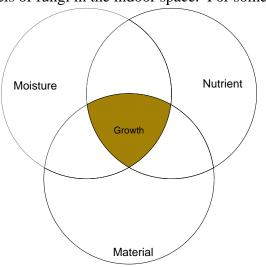
Figure 2 illustrates the combination of moisture and nutrients required for microbial growth on a material. Sufficient nutrients for growth may be provided by the material itself or through the accumulation of dust on or in the material. For wallboard, some of the component(s) of the

the nutrients. When sufficient nutrients are available, the ultimate determinate for microbial growth is availability of water. The more hygroscopic a material (i.e., wallboard) is, the more impact on the overall hygoscopicity the surface treatments may have.

This test plan addresses three specific characteristics of mold resistant building material: (1) mold resistance, (2) emissions of VOCs and aldehydes, and (3) moisture content. Mold resistance is the critical measurement, so the T/QAP is focusing on mold resistance. Moisture content and emissions of VOCs and aldehydes are ancillary tests; information can be found in Appendix A and B, respectively. Other characteristics, such as fire resistance, are important and should be considered by users of the products, but are beyond the scope of this test plan.

A building is not a sterile environment, nor should it be. In fact, a building is frequently a reservoir for microorganisms. While many different types of microorganisms occupy indoor spaces, it is well-recognized that fungi can colonize and amplify on a variety of building materials if sufficient nutrients and moisture are present. These contaminated materials are known to be important indoor reservoirs. Fungal growth on natural and fabricated building materials can be a major source of respiratory disease in humans.

Table 1 provides information on common environmental fungi that have been isolated from contaminated wallboard.



wallboard itself may provide the necessary nutrients, but wallpaper and paint can also provide

Figure 2. Diagram illustrating the conditions required for fungal growth on a material.

Table 1. Common lungi lound on wetted wandoard.				
Organism	Materials That Support Growth			
Acremonium spp.	Colonizes damp wallboard			
Alternaria spp.	Grows indoors on a variety of substrates			
Aspergillus spp.	Colonizes damp wallboard			
Chaetomium spp.	Commonly found on damp wallboard paper			
Cladosporium spp.	Colonizes continuously damp wallboard			
Epicoccum spp.	Colonizes continuously damp wallboard			
Fusarium spp.	Colonizes continuously damp wallboard			
Penicillium spp.	Commonly found on damp wallpaper			
Stachybotrys spp.	Colonizes continuously damp wallboard			
Trichoderma spp.	Grows well on damp wallboard paper			

				_	
Table 1	Common	fungi	found on	wetted	wallboard.
Table L.	Common	Tungi	iouna on	wittu	wannoaru.

Mold resistance testing will be performed following the guidelines outlined in ASTM 6329-98<sup>9</sup>. This method was developed as part of a more comprehensive project to apply indoor air quality engineering to biocontamination in buildings. One of the primary goals was to provide a scientific basis for studying indoor air biocontaminants. Available methods from ASTM, AATCC, UL, and others for evaluating the resistance of a variety of materials to fungal growth were surveyed at the initial stages of that project. Although the basic principals were similar, a major concern was the way growth on the different materials was evaluated. Although in some cases, quantitative methods for inoculation were employed, none assessed growth as the endpoint quantitatively. The strategy was to develop a method that would provide a quantitative endpoint for growth in a well-controlled environment and to improve repeatability and comparability. The method has been successfully used to evaluate fungal resistance on a variety of materials including ceiling tiles and HVAC duct materials <sup>10,11,12,13</sup>.

# A6: ESTE Testing - Description and Schedule

The ESTE testing for mold-resistant gypsum board will be performed under the U.S. EPA's quality system where the goal is to ensure that environmental programs and decisions are supported by data of the type and quality needed and expected for their intended use, and that decisions involving environmental technology are supported by appropriate quality-assured engineering standards and practices. The implementation of the EPA Quality System is based on a graded approach, meaning that quality systems for different organizations and programs will vary according to the specific objectives and needs of the organization. EPA requires that contractors performing research or testing for EPA have both a quality management plan<sup>1</sup> and a quality assurance project plan (QAPP) (or test plan) – this document.

# The ETV management plan states:

The quality system for the overall ETV program seeks to be consistent with industry consensus standards. Each verification organization shall implement a valid and approved quality system. The Agency's required quality system for cooperative agreements and contracts is ANSI/ASQC E4. Each verification test will be performed according to planned and documented, pre-approved test/QA plans. All technical statements in ETV verification reports shall be supported by the appropriate data<sup>14</sup>.

ETV requires that all test plans, data and results be reviewed by both the company (in this case, RTI) and the EPA project quality managers. In addition, both RTI and EPA perform audits of the

RTI ESTE quality system and how the testing is being performed and how ETV quality management performs.

EPA uses the data quality objective (DQO) process as defined in Guidance on Systematic Planning using the Data Quality Objectives Process (QA/G-4)<sup>15</sup>. The DQO process is used to establish performance or acceptance criteria, which serve as the basis for designing a plan for collecting data of sufficient quality and quantity to support the goal of a study<sup>15</sup>. DQOs may include specific values for accuracy and precision, but they will often provide one value that incorporates both entities.

EPA and ETV strongly support the use of international consensus standards, however, recognizing that a laboratory will need to supplement standards with their own SOPs. The following sections indicate which standards are used in developing this QAPP/test plan. This document is written in a format defined by EPA that includes sections not found in a standard test method. This document will indicate sections and document information when procedures are being used from those documents.

# A6.1: Description of Testing

The ESTE verification test includes separate tests to be performed on each gypsum board submitted for testing:

- Mold resistance testing.
- VOC and aldehyde emissions testing, and
- Moisture content

Testing for VOC and aldehyde emissions and moisture content are ancillary tests.

The testing consists of the three steps summarized below:

- Acquiring the mold-resistant gypsum board products for testing,
- Performing the testing of the boards, and
- Preparing verification reports and summary statements.

Each of the three tests is discussed separately. The moisture test can be found in appendix A. The VOC and aldehyde emissions test can be found in appendix B.

# A6.1.1: Identification and Acquisition of Mold-Resistant Gypsum Board

The companies will provide one bundle (two sheets) of their gypsum board to RTI for mold resistance and moisture content testing. Tricia Schwartz will be the custodian at RTI and will be responsible for storage, labeling, etc. of the products. She is responsible for documentation showing chain of custody. Each piece of wallboard will be labeled as will the samples cut from it. The products will be stored in the Building 11, Bay 1 storage room at RTI until used and will be retained until the verification report for the product is approved. All products will be logged in upon receipt and will be checked out for testing, then the unused portion will be returned to the storage room when testing is complete.

A minimum of four 12" x 12" squares will be shipped to AQS for VOC testing using AQSapproved packaging. AQS will follow their own procedures for sample collection and handling (AQS Method, Sections 5.3-5.9 – Appendix C.

# A6.1.2: Performance of Mold Resistance Testing

Mold resistance testing will be performed following the guidelines outlined in ASTM 6329-98<sup>9</sup>. The specifics of the test and the related SOPs are discussed in further detail in Section B1.2. In overview, the test organisms are inoculated by pipette directly onto the surface of each wallboard piece in sufficiently high numbers to provide an adequate challenge, but at a level that is realistic to quantitate. The tests will run for 12 weeks. Within the 12 weeks of the test, four test days, Day 0, Week 1, Week 6, and Week 12 will be evaluated. Day 0 will provide the baseline inoculum level. A sufficient number of test pieces will be inoculated simultaneously for all four test days. All of the pieces for one material and one test organism will be put in the same static chamber. Two test organisms, *Stachybotrys chartarum* and *Aspergillus versicolor* will be used. The chambers will be set to 100% ERH for the tests with *S. chartarum* and at 85% for *A. versicolor*. On each test day (including day 0), five replicates of the test material pieces are removed from the chamber, placed in sterile buffer, and extracted by shaking. The resulting suspension of eluted organisms is plated and microbial growth on materials quantitated by manually enumerating colony-forming units (CFU),

The numbers of CFU eluted on test days week 1, 6, and 12 and compared to the baseline at Day 0. The numbers of CFU are expressed as  $log_{10}$ . The results will be reported as the log change in CFUs between Day 0 and Week 1, Day 0 and Week 6, and Day 0 and Week 12.

# A6.1.3: Preparation of Report

The final step is to complete the verification report and verification statement for each product tested and submit them to EPA for review.

# A6.2: Schedule

The verification will begin when the T/QAP is approved by EPA and will continue at least until all three initial products are tested. The vendors who have taken part in the test plan development will be requested to submit their products while the T/QAP is in EPA review. Testing may continue beyond that if there are additional products to test.

# A7: Data Quality Objective and Criteria for Measurement Data

Data quality objectives (DQOs) are qualitative and quantitative statements designed to ensure that the type, quality, and quantity of data used are appropriate for the intended application. The DQO for the critical measurement, quantitation of fungal growth on an individual test date, is found in Table 2.

			DQO	
Test	Parameter	Precision	Accuracy	Completeness
Mold Resistance	Quantitation of fungal growth on an individual test date	± 5-fold difference	10% of the plates will be counted by a second operator. ± 20% agreement between the operators	100%

Table 2. Data Quality Objectives

All RTI and AQS data will be reviewed for accuracy (correctness) and reasonableness. If the results are deemed unreasonable (e.g., internally inconsistent), they will be discarded, the procedures reviewed, and the test repeated if necessary. Data points that are analyzed and determined to be obvious outliers will be discarded without requiring the entire test to be repeated.

# A8: Special Training Requirements/Certification

There are no specialized certification requirements specified for these tests. The RTI Project Manager will be responsible for overseeing all work and ensuring that RTI personnel are fully trained in each operation for the microbial and moisture testing. RTI personnel working with the mold resistance testing are trained to perform all of the required procedures, including determining microbial growth. Under their ISO 9001-2000 quality management system, the AQS quality assurance report (Appendix C) documents that all staff are trained and knowledgeable for the VOC testing they will perform.

# **A9: Documentation and Records**

This section identifies the documents and reports to be generated as part of the verification program and the information to be included in the verification reports. A description of the data management system established for this task is presented in Section B.10.

Requirements for record keeping and data management for the overall program are found in the Quality Management Plan (QMP) for the Verification Testing of Air Pollution Control Technology, Revision 2.2<sup>1</sup>, including Table 5-1, Records Management Responsibilities for ETV. All RTI microbiology laboratories (ML) SOPs and TOPs (Technical Operating Procedures) are maintained on file at RTI. Access to these files is permitted on-site at RTI and will be available for review during internal and external TSAs. AQS will make their SOPs available for review.

# **A9.1: Laboratory Documentation**

The culturable test organism counts or CFUs from each test piece on each test day will be entered in the project notebook or recorded by a computer. If recorded to a computer, the file will be saved to the hard drive and later copied to a CD or shared directory for backup.

# A9.2: QA Reports

The RTI QAM will perform an internal TSA based on the approved T/QAP during the first month of verification; this is considered suitable because this testing program is using well-known measurement systems components. A report will be prepared for the Project Manager within 15 days of completion of the audit.

The RTI QAM will perform an ADQ of all RTI and AQS data. As part of the ADQ, RTI will also review AQS QA results and verify that AQS has met all QA/QC requirements for this project. RTI will provide a report of the ADQ to EPA with the verification report for review.

RTI will cooperate with audits performed by the EPA Project Officer, EPA QAM, or their designee insofar as resources permit. EPA will perform a TSA for this project.

# **A9.3: Reporting**

After the completion of tests, the control test data, sample inventory logs, calibration records, and certificates of calibration will be stored in the laboratories. Copies of these will be made for RTI and EPA QA. Calibration records will include such information as the instrument being calibrated, raw calibration data, calibration equations, analyzer identifications, calibration dates, calibration standards used and their traceabilities, identification of calibration equipment used, and the staff conducting the calibration. Final reports of self-assessments and independent assessments [i.e., technical systems audits, performance evaluations, and audits of data quality (TSAs and ADQs)] will be retained as required by ETV and RTI. Each verification report will contain a QA section, which will describe QA activities and the extent to which test data comply with DQOs.

# **A9.4: Verification Reports**

Verification reports will be prepared by RTI personnel and reviewed by RTI's Project Manager and QAM prior to submittal to the EPA Project Officer for review.

There shall be a general discussion of the test process giving dates and other information as appropriate.

The microbial resistance will be reported as the log change in CFUs between the start date and each of the four test dates. Each test organism will be reported separately.

The DQO will be reported. The results of the ancillary moisture and emissions tests will also be included.

# A9.4.1: Environmental Sustainability Criteria

A section of the verification report and the verification statement will focus on environmental sustainability, with information provided by the vendor.

The verification organization will work with the vendor to estimate impacts on solid waste disposal from replacing wallboard less frequently, using the test results for microbial resistance. If possible this should be done quantitatively with uncertainty estimates.

The gypsum board vendor will supply the following:

- Information regarding the chemicals or other product characteristics that engender the product microbial resistant.
- For chemical additives that are claimed to confer microbial resistance, the vendor shall provide the identity of the chemical and a summary of toxicity information relative to the chemical. The quantity of the chemical used in the product wallboard shall also be provided. It is expected that this information will come from the MSDS sheet, which can be included as an appendix to the report.
- Additional information relative to the environmental sustainability of the product such as recyclability/reusability of the product and disposability of the product and use of renewable resources or other criteria the vendor deems relevant to the environmental sustainability of the product.

### SECTION B: MEASUREMENT/DATA ACQUISITION

#### **B1: Test Design**

Mold resistance testing will be based on ASTM D6329-98<sup>9</sup>, "Standard Guide for Developing Methodology for Evaluating the Ability of Indoor Materials to Support Microbial Growth Using Static Environmental Chambers".

#### **B1.1** Static Chambers

Acrylic desiccators will serve as the static environmental chambers. The desiccators are sealed so there is no air exchange and serve as good static chambers. The chamber humidity will be maintained through the use of saturated salt solutions (ASTM E104-02<sup>16</sup>) or sterile water. Temperature control is externally controlled and maintained at room temperature. Prior to use, the chambers will be decontaminated following ML SOP #017 (Standard Operating Procedure for the Decontamination of Humidity Chambers). The chambers will be characterized following ML SOP #005 (Standard Operating Procedure for the Characterization of Relative Humidity Chamber) once prior to initiation of the testing.

The chambers will be set to 100% ERH for the tests with *Stachybotrys chartarum* and at 85% for *Aspergillus versicolor*. The ERH in each chamber will be monitored with a hygrometer.

### **B1.2** Test Organisms

Selecting the "correct" test organism is critical to any test; therefore selection criteria were developed. The selection criteria used to choose the appropriate test organisms for this study were:

- (1) the reasonableness or likelihood of the test material being challenged by that particular organism when in actual use, and
- (2) that they cover the range of ERHs needed to bracketing the ERHs where fungal growth can occur on building materials.

While exposure to many of the fungi can be considered problematic, none are as controversial as exposure to *Stachybotrys chartarum*<sup>17,18</sup>. There are numerous reports demonstrating an association between exposure to *S. chartarum* and adverse respiratory heath effects; but none is as compelling as the paper simply entitled *Stachybotrys* by Etzel<sup>19</sup>, which reviews what has been learned about *Stachybotrys* and its association with pediatric pulmonary disease. Pulmonary hemosiderosis among infants was reported in Cleveland, OH, in the early 1990s (Etzel<sup>20</sup> et al.) although the findings were not sufficient to support an association between *Stachybotrys chartarum* on wallboard and the disease (Montana<sup>21</sup> et al., Etzel<sup>22</sup> et al., CDC MMWR<sup>23</sup>). In another study, mycotoxin produced from *S. chartarum* on wetted wallboard has been found to be water-soluble and was toxigenic in vitro (Black<sup>24</sup> et al.).

Two fungi will be used as test organism, *Aspergillus versicolor* and *Stachybotrys chartarum*. Both are from the RTI culture collection (CC). The CC number for *S chartarum* is 3075 and was received from EPA NERL. *A. versicolor* is CC #3348, and it is a field isolate. Prior to initiation of the testing, their identification will be confirmed by standard techniques. *Aspergillus versicolor* is recommended because it is a xerophilic fungus and capable of growing at lower relative humidities. *Stachybotrys chartarum* is recommended because it requires high levels of available water to grow and has been associated with a number of toxigenic symptoms. *A*. *versicolor* is frequently reported as a causative agent of hypersensitivity pneumonitis and has been isolated from a number of problem buildings. Both have been reported as growing on gypsum wallboard.

### **B1.3** Sample Preparation and Inoculation

Small (at least 4 cm x 4 cm), replicate pieces of test mold resistant wallboard will be prepared and inoculated. To minimize error and demonstrate reproducibility, five pieces of each sample type will be processed on each respective sampling day. Because there are four test dates, a minimum of 20 pieces will be prepared simultaneously. Each piece is placed on a separate labeled sterile petri dish

Fungi media, cultures, spore isolation, suspension, quantitation, and inoculation on the test wallboard will follow: ML SOP #001 (Standard Operating Procedure for Media Preparation – Dehydrated); ML SOP #002 (Standard Operating Procedure for the Preparation of Sterile Water); ML SOP #003 (Standard Operating Procedure for the Preparation of Sterile Buffer); ML SOP #009 (Standard Operating Procedure for the Quantitative Evaluation of Microorganisms); ML SOP #012 (Standard Operating Procedure for the Quantitation of Viable Spores in Suspension Preparation); and ML SOP #058 (Standard Operating Procedure for Direct Inoculation of Materials with a Spore Suspension). A review of those steps follow.

The fungi challenge suspensions are prepared by inoculating the test organism onto solid agar media, incubating the culture at room temperature until mature, wiping organisms from the surface of the pure culture, and eluting them into sterile18-Mohm distilled water to a known concentration to serve as a stock solution. The organism preparation is viewed microscopically to verify purity of spores (absence of hyphae). The suspension is diluted in sterile 18-Mohm distilled water if needed to a concentration of approximately  $10^5 - 10^6$  CFU/mL. The test pieces are inoculated (usually with 5 10µL spots in an X configuration) by pipet onto the surface of the wallboard test piece and allowed to dry in the biosafety cabinet. The goal is to load each of the individual test pieces with approximately  $10^3$  to  $10^5$  CFU/piece. The fungi spore suspension is quantified on appropriate media to enumerate the inoculum.

On each test day (including day 0), the test pieces will be removed from the static chamber, placed in approximately 30 mL sterile buffer, and extracted by shaking using a vortex or wrist action shaker. The extract is diluted if needed and plated on agar media to determine CFU.

## **B2: Sampling Methods Requirements**

The mold resistance sampling method requirements and critical dimensions and configurations of the test chamber are specified in Foarde<sup>25</sup> et al. All sampling methodology will comply where appropriate. Static chambers are maintained in accordance with ML SOP #005 (Standard Operating Procedure for the Characterization of Relative Humidity Chamber), ML SOP #009 (Standard Operating Procedure for the Quantitative Evaluation of Microorganisms), ML SOP #017 (Standard Operating Procedure for the Decontamination of Humidity Chambers), and ML SOP #058 (Standard Operating Procedure for Direct Inoculation of Materials with a Spore Suspension). Where used, all equipment will be calibrated and operated according to the manufacturer's specifications.

Each SOP has its own QUALITY ASSURANCE/QUALITY CONTROL section, which includes the QC checks, which are available at RTI. As one example, ML SOP #001 Standard Operating Procedure for Media Preparation – Dehydrated, the QC checks are as follows:

- If balance has an internal calibration program, that may be used, or a check weight can be used as first and last weighing. Refer to the procedures outlined in the appropriate user's manual for more specific calibration instructions if needed.
- Balances are professionally calibrated annually.
- If the Manostat is used, the desired volume of media dispensed is checked with a graduated cylinder or tube before filling plates.
- Incubate plates at room temperature (on counter) for at least 3 days (no more than 5) prior to use so that contaminated plates may be identified (appearance of any colonies) and disposed of in an appropriate manner. Media for bacteria may be used if necessary after 1 day.
- Inoculate several plates (~2 out of 100) with a loop of the appropriate organism to prove the media's ability to sustain growth (see supervisor to ascertain incubation time and temperature).
- Record the reaction in the media preparation notebook.
- After quality control tests have been completed, store bagged and labeled plates upside down in a refrigerator (2-5 °C).

# **B3:** Sample Handling and Custody Requirements

Sampling methods and laboratory procedures are described in specific laboratory SOPs. These SOPs address any anticipated failures and the methods that will be employed to overcome these failures. Most of the methods are well-known sampling methods; therefore, sampling failures are not anticipated. Any additional project-specific considerations will be addressed and included in an updated SOP. Supporting measurements, such as temperature, relative humidity or atmospheric pressure, will be recorded in laboratory data logs, run sheets or notebooks.

# **B4: Analytical Methods Requirements**

The analytical method requirement for the mold resistance in static chamber test is described in ASTM D6329-98<sup>9</sup>. The requirements for biological testing are described in the appropriate ML SOPs.

# **B5: Quality Control Requirements**

The static chambers are decontaminated and cleaned in accordance with ML SOP #017 (Standard Operating Procedure for the Decontamination of Humidity Chambers). All media and reagents are handled as outlined in ML SOP #001 (Standard Operating Procedure for Media Preparation – Dehydrated) and ML SOP #003 (Standard Operating Procedure for the Preparation of Sterile Buffer). All laboratory surfaces are disinfected using ML SOP #023 (Standard Operating Procedure for Regular Disinfection of Laboratory Surfaces). All laboratory equipment including the autoclave sterilizer is verified using ML SOP #019 (Standard Operating Procedure for Maintenance and Record Keeping of Laboratory Equipment).

### **B6:** Instrument/Equipment Testing, Inspection, and Maintenance Requirements

RTI ML instrument maintenance is done in accord with the ML's SOPs.

## **B7: Instrument Calibration and Frequency**

Calibration will be performed in accordance with the manufacturer's recommendations or annually. Recommended instrument calibration frequencies are provided in the respective SOPs and/or manufacturer's manuals. Pipettes will be calibrated gravimetrically following ML SOP #013 (SOP for Pipet Calibration).

Calibration for chemical analysis instruments are defined in the AQS test method Section 6 (Appendix C).

### **B8:** Inspection/Acceptance Requirements for Supplies and Consumables

Chemicals, supplies, and other consumables will be purchased from sources that have provided high-quality products to the laboratory in the past. Materials such as growth media will be purchased from a single source to help ensure uniformity throughout the duration of the project. All supplies will be inspected by the lab personnel. RTI's purchasing department will assist with the return of any equipment or materials that do not meet project requirements. Items will be NIST traceable when possible.

## **B9:** Data Acquisition Requirements (Non-direct measurements)

No types of data are needed for project implementation or decision making that would be obtained from non-measurement sources such as computer databases, programs, literature files, or historical databases.

Manual methods of primary data acquisition (e.g., visual CFU counting) are described in ML's SOPs, while automated data acquisition equipment (e.g., balances and environmental controls) is checked using procedures recommended by the manufacturer. Procedures for screening and verifying manually entered data are used to reduce input errors to a minimum through different members of the staff double checking each other. Non-experimental data, such as an MSDS, will be included in the project notebook and a copy maintained in the RTI Project Manager's project file.

AQS follows data acquisition procedures as specified in its test plan and ISO 9001 plan.

## **B10: Data Management**

Guidelines for data management in the ML include the description, location, format, and organization of all types of records. The RTI Project Manager will oversee all data management activities. This section identifies the activities and processes planned for documenting the traceability of the data, calibrations, and information in the verification report. Corrections to manual entries are made on the same line (where possible), and are initialed and dated.

## **B10.1: Data Recording**

Data for this task will be collected either by computer or by manual (handwritten) entries. Observations and records (e.g., sample description and collection information) will be recorded manually in lab notebooks kept exclusively for this task.

# **B10.2:** Data Analysis

Analysis will be performed as defined in ASTM D6329-98, Section 12.3.3.

# **B10.3: Data Storage and Retrieval**

Laboratory notebooks containing manually recorded information and data output generated from instrumentation will be stored in the custody of the project leader for the duration of the project. Access to the notebooks is controlled; any changes made are initialed and dated, thus maintaining a good audit trail.

Spreadsheet files including raw and calculated data will be stored on computers. The files will be downloaded to a network server which is backed up nightly. Access to the files are controlled, with file names indicating date and initials of person creating file, thus maintaining a good audit trail.

Following ETV policy, data files will be archived for 10 years following the end of the project and reports will be kept in perpetuity. The records will not be destroyed without written approval from EPA.

#### SECTION C: ASSESSMENT/OVERSIGHT

#### **C1:** Assessments and Response Actions

#### C1.1: Audits

RTI will be subject to both external and internal audits as specified in the ETV QMP<sup>1</sup>, especially Table 9-1, ETV Assessments. A subset of that table is shown below, Table 3. Audits based on this test/QA plan include Technical System Audits (TSAs). RTI and AQS raw and summary data are subject to Audits of Data Quality (ADQs). An external TSA will be conducted by EPA or a designated representative. Other external audits may be performed. The auditor(s) will document their findings and note where corrective actions are necessary. The auditor(s) will distribute audit reports to those listed in Section A3 as well as to the supervisor whose laboratory was audited. RTI will provide the QA reports, including the ADQ report, to EPA with the verification report for review.

Assessment Tool	Assessors	Subject of	Minimum	Reason for	Report Reviewed by
		Assessment	Frequency	Assessment	
Technical	Self	Test/QA plan	Self	Assess	EPA Project Officer
Systems Audits	RTI QAM		Once per technology	technical	
			evaluation	quality of	EPA QAM
	Independent			evaluations	
	EPA QAM		Independent		RTI Project Manager
			Once per year, as		
			applicable		
Audits of Data	Self	Raw data and	Self	Assess data	EPA Project Officer
Quality	RTI QAM	summary data	At least 10% of the	calculations	
		-	data in each	and reporting	EPA QAM
	Independent		technology		
	EPA QAM		evaluation		RTI Project Manager
			Independent		
			Each technology		
			evaluation, as		
			applicable		

### Table 3. RTI's ETV Assessments

## **C1.2:** Corrective Actions

Technical personnel will have the direct responsibility for ensuring that whenever accuracy or bias is outside the limits of the DQOs for the critical measurements that corrective actions are taken. If procedures are found to be faulty, corrective action will also be taken.

Corrective actions include:

- Problem identification;
- Attempting to find the cause;
- Attempting immediate steps to remedy the situation (if possible);
- Reporting or documenting the problem;
- Planning for corrective action (if major repairs are needed);
- Checking that the problem was corrected;

- Documenting the corrective actions taken; and
- Recommending changes to instruments, SOPs, etc., to avoid similar future occurrences.

The RTI QAM and Project Manager will be jointly responsible for proper documentation of corrective actions. Minor corrective actions are to be recorded in the laboratory notebooks. Major problems will be addressed as outlined above. All corrective actions will be noted in the verification report. Depending on the time and expense involved with necessary corrective actions, it will be necessary to consult the Program Manager or the sponsor before implementing any changes in the planned activities.

### **C2:** Reports to Management

The RTI Project Manager will notify the EPA Project Officer when testing under this project is being conducted. The RTI Project Manager will submit verification reports, as well as data, to the RTI QAM. The RTI QAM will submit reports of all technical assessments to the RTI Project Manager. The RTI Project Manager will submit verification reports to the EPA Project Officer.

TSA and ADQ reports will be sent to the EPA Project Officer and QAM.

#### D1: Data Review, Validation, and Verification Requirements

The verification is acceptable if all the measured parameters fall within the DQO limits described in Table 2. The test operator and analyst are responsible for checking that all measured parameters fall within prescribed limits before continuing testing.

### **D2: Validation and Verification Methods**

Both the test operator and the test analyst will verify that the test data have been correctly entered and processed and that all manual calculations are correct. They will verify all newly developed or modified software, including spreadsheets for correctness before using the software to process project data.

Each verification report will be reviewed by the RTI QAM for compliance with the applicable method and for the quality of the data reported.

The RTI QAM will check for the following:

- Data completeness,
- Initial and continuing calibrations, and
- QC reference and internal standards.

## D3: Reconciliation with Data Quality Objectives

Each ETV verification report will present the critical and relevant ancillary measurements.

The program manager will be responsible for reconciling data sets with the DQOs. She will work with the QAM for the mold resistance testing and with RTI technical staff and AQS for the moisture and emissions testing. This will be done during the ADQ and will be reported in the ADQ report.

Actual data quality will be compared with the DQO specified in Section A7; if the data quality meets or exceeds the objectives and verification specifications have been met, the test data will be considered acceptable. If exceptions are identified, the issues will be investigated for impact on the credibility of the data, the EPA QAM will be consulted, and the verification results disposed of on the basis of this careful consideration. If the impact is not significant, the data will be reported with indication of the exceptions.

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### **RTI Operating Procedures Referenced in the Test/QA Plan**

ML SOP #001	Standard Operating Procedure for Media Preparation – Dehydrated
ML SOP #002	Standard Operating Procedure for the Preparation of Sterile Water
ML SOP #003	Standard Operating Procedure for the Preparation of Sterile Buffer
ML SOP #005	Standard Operating Procedure for the Characterization of Relative Humidity
	Chamber
ML SOP #007	Standard Operating Procedure for the Gravimetric Determination of Moisture
	Content
ML SOP #009	Standard Operating Procedure for the Quantitative Evaluation of
	Microorganisms
ML SOP #012	Standard Operating Procedure for the Quantitation of Viable Spores in
	Suspension Preparation
ML SOP #013	Standard Operating Procedure for Pipet Calibration
ML SOP #017	Standard Operating Procedure for the Decontamination of Humidity Chambers
ML SOP #019	Standard Operating Procedure for Maintenance and Record Keeping of
	Laboratory Equipment
ML SOP #023	Standard Operating Procedure for Regular Disinfection of Laboratory Surfaces
ML SOP #058	Standard Operating Procedure for Direct Inoculation of Materials with a Spore
	Suspension

Appendix A Moisture Testing

### **Impact of Moisture on Building Materials**

It is well established that molds can colonize and amplify on a variety of building materials if sufficient nutrients and moisture are present. Commonly, sufficient nutrients are available and water is usually the growth factor most limiting the establishment and growth of microbial populations. Sufficient moisture for growth may become available through water incursion from leaks and spills, condensation on cold surfaces, or absorbtion or adsorption directly from the indoor air. The amount of water required is not large, and materials that appear dry to cursory inspection may be capable of supporting microorganism growth. Because moisture is so important, knowing the amount of available moisture in a material is critical to preventing and controlling microbial growth and amplification on materials.

### **Performance of Moisture Testing**

Moisture testing will also be performed following the guidelines outlined in ASTM 6329. Two types of measurements are commonly used to evaluate building material moisture: most engineers think in terms of moisture content (MC) and many microbiologists utilize water activity ( $a_w$ ). MC, defined as mass of water per unit mass of dry material, is measured gravimetrically (West and Hansen<sup>26</sup>; Foarde<sup>27</sup> et al.). It is a bulk measurement of the water in a sample of the material. The  $a_w$ , primarily used to relate the water content of foods to the ability of microorganisms to grow on them, is defined as the equilibrium RH (ERH) above a sample of a material, divided by 100 (Pitt<sup>28</sup>).

In the test described in this document, the ERH is the RH in a closed chamber containing a material sample after the material and the air in the chamber have reached water equilibrium. Therefore, the  $a_w$  of the material that has been equilibrated in a closed chamber having an RH of 94% is 0.94. Corry<sup>29</sup> stated that  $a_w$  is the proportion of "available water for biological reactions." It is a useful laboratory measurement when RH conditions are known to be at equilibrium.

For porous materials such as wallboard, MC and ERH (or  $a_w$ ) are related through the material's water adsorption isotherm, and different relationships are obtained for different materials. In this test, the MC for each test material will be determined at four ERHs, 100%, 94%, 85% and 62%. There is no need to test lower ERHs as microbial growth cannot occur below ~ 60% ERH.

For microbial growth, the a<sub>w</sub> or ERH is the important number as it indicates how much water is available for growth. The MC will be determined for each test material, but will be reported only as a reference value. This reference value will provide two pieces of information. First, the actual MC at the time of the test. This may be useful for comparison purposes if other samples of the same material are tested at a later date. Additionally, while the microbial resistance testing will be performed with the test material at ERH, in buildings the RH of a space and the materials in the space are not at equilibrium. Therefore, another measure such as MC would be important.

## **Test Design for Moisture Testing**

In addition to ASTM D6329, ML SOP #007 (Standard Operating Procedure for the Gravimetric Determination of Moisture Content) will be followed. The details are described below.

Replicate small (at least 4 cm x 4 cm) pieces of the test material will be used. Pieces will be

placed on sterile dishes on the shelves in the static chamber. Equilibration time will depend upon both the material to be tested and the chamber relative humidity selected for the test and will be determined for each material prior to testing. Equilibration will be defined as when the bulk moisture content of the material reaches a constant value. A calibrated analytical balance will be used.

At least five chambers will be used. One each will be set at 100%, 94%, 85% and 62% ERH. The fifth chamber will contain a desiccant for a near 0% RH baseline comparison.

### **Data Analysis for Moisture Testing**

Analysis will be performed as defined in ASTM D6329, Section 8.3.2.

#### **Appendix B. Emissions Testing**

#### Impact of Emissions of VOCs and Aldehydes from Building Materials

Volatile organic compounds (VOCs) and aldehydes are of a concern for indoor air quality because of adverse health effects. Many items in buildings may emit VOCs and aldehydes, including paints and lacquers, paint strippers, cleaning supplies, pesticides, building materials and furnishings, office equipment such as copiers and printers, correction fluids and carbonless copy paper, graphics and craft materials including glues and adhesives, permanent markers, and photographic solutions. Manufacturers of these items have been working since the early 1990s (or earlier) to reduce emissions from their products. EPA has sponsored research and testing on low emission products. Under an earlier RTI developed a test method for measuring VOC and aldehyde emissions from commercial furniture<sup>30</sup>.

This test plan includes measurement of emissions of aldehydes and VOCs from gypsum board under conditions designed to approximately simulate product use. Formaldehyde and total volatile organic compounds (TVOC) can be measured in addition to a range of other aldehydes and individual VOCs. Emissions levels are determined by placing a piece of the board into a small environmental test chamber under specified test conditions then measuring chamber air concentrations of aldehydes and VOCs at selected time intervals. Product-specific emission factors are calculated from the chamber air measurements. The method provides a standard test to reproducibly and accurately measure emissions from gypsum board under controlled laboratory conditions.

Chemical testing of VOCs and aldehydes will be performed by Air Quality Sciences (AQS) as a service, not as a subcontract. AQS will use the "Method for the Evaluation of Emissions from Gypsum Board Using Environmental Chambers.<sup>31</sup>" (AQS method – Appendix C) AQS was a subcontractor to RTI for the Indoor Air ETV Pilot and participated in round-robin volatile organic compound (VOC) testing. The company was founded in 1989 by Marilyn Black to provide chemical and biological services to commercial and government clients. AQS provides emissions testing of consumer products, construction materials, and interior furnishings for chemical and particle releases to indoor environments and verification of the data for consumer and regulatory acceptance and risk evaluation. Their laboratories are ISO 9001:2000 registered for quality management. AQS publishes a quarterly quality assurance report with details of their quality system and the latest results for key performance measurements.

#### **Performance of Emissions Testing**

The emissions testing will be performed by AQS using their "Method for the Evaluation of Emissions from Gypsum Board Using Environmental Chambers<sup>31</sup>" (Appendix C). Testing follows the guidance of ASTM D5116-06<sup>32</sup>, "Standard Guide for Small Scale Environmental Chamber Determinations of Organic Emissions from Indoor Materials/Products"; ASTM, D6670-01<sup>33</sup>, "Standard Practice for Full-Scale Chamber Determination of Volatile Organic Emissions from Indoor Materials/Products"; ASTM, D6670-01<sup>33</sup>, "Standard Practice for Full-Scale Chamber Determination of Volatile Organic Emissions from Indoor Materials/Products"; and RTI's ETV protocol for EPA<sup>30</sup>, "Large Chamber Test Protocol for Measuring Emissions of VOCs and Aldehydes From Commercial Furniture". Other standards are used for measurement and analysis as described in the AQS test method. RTI will send AQS samples to be tested. A minimum of four 12" x 12" squares will be shipped to AQS using AQS-approved packaging. AQS will follow their own procedures for sample collection and handling (AQS Method, Sections 5.3-5.9 – Appendix C). A sample is

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placed into an environmental chamber. Measurements are taken over time for VOCs and aldehydes. Gas chromatography/mass spectrometry (GC/MS) is used to determine the quantity of VOCs and reverse-phase high-performance liquid chromatography (HPLC) with ultraviolet (UV) detection is used for aldehydes.

### **Test Design for Emissions Testing**

The testing will be performed according to the AQS method, Section 6, Chamber Testing Protocol (Appendix C). Emissions levels are determined by placing the test objects into an environmental test chamber under specified test conditions then measuring chamber air concentrations of aldehydes and VOCs at selected time intervals. Product-specific emission factors are calculated from the chamber air measurements.

Aldehydes in chamber air samples are collected on silica gel cartridges coated with 2,4-dinitrophenylhydrazine (DNPH). The DNPH-aldehyde derivatives on the cartridges are eluted with acetonitrile then analyzed by high performance liquid chromatography (HPLC) with ultraviolet (UV) detection.

VOCs in chamber air samples are collected on sorbent cartridges (tubes). VOCs trapped on the cartridges are thermally desorbed then analyzed by gas chromatography/mass spectrometry (GC/MS). Results of these analyses will be reported as  $mg/m^3$  (or  $\mu g/m^3$ ) and may be used to estimate concentrations in chamber air samples.

Sampling will be performed according to the AQS method, Section 6 (Appendix C), as described in Section B1.1 of this document. The AQS test method specifies guidelines in Section 5, Sample Collection, Handling, and Product Tracking.

#### **Analysis for Emissions Testing**

Chemical analysis will be performed according to the AQS Method, Section 6 (Appendix C).

For individual VOCs, the analytical methods are based on ASTM D6196-03<sup>34</sup>, "Standard Practice for Selection of Sorbents, Sampling, and Thermal Desorption Analysis Procedures for Volatile Organic Compounds in Air". AQS also references EPA methods T017<sup>35</sup>, "Determination of Volatile Organic Compounds in Ambient Air Using Active Sampling onto Sorbent Tubes" and TO-1<sup>35</sup>, "Determination of Volatile Organic Compounds in Ambient Air Using Tenax Adsorption and Gas Chromatography/Mass Spectrometry".

The analytical methods for formaldehyde, and other low molecular weight aldehydes are based on ASTM D5197-03<sup>36</sup>, Standard Test Method for Determination of Formaldehyde and Other Carbonyl Compounds in Air (Active Sampler Methodology)".

Data analysis will be performed as defined in the AQS method, Section 6 (Appendix C).

# **Quality Assurance and Control for Emissions Testing**

QC requirements for emissions testing will be performed according to the AQS Method, Section 6 (Appendix C) and include the following:

- 1. Air change: 1.0ACH +/- 5.0%
- 2. Mixing: +/- 5.0%
- 3. Air Tightness: <0.03 ACH
- 4. Recovery\*: 100% +/- 20%

\*recovery is based on average recoveries from Toluene, Decane, and Formaldehyde. The RTI program manager and QAM will oversee the AQS QC process.

AQS follows acquisition procedures as specified in their ISO 9001 plan. Under their ISO 9001-2000 quality management system, AQS documents that all staff are trained and knowledgeable for the work they perform. Calibration for chemical analysis instruments are defined in the AQS test method Section 6 (Appendix C).

All AQS data will be reviewed for accuracy (correctness) and reasonableness. In addition, AQS raw and summary data are subject to ADQs. AQS shall provide RTI sufficient documentation that RTI and EPA will be able to perform audits. If there are proprietary SOPs or other documents, an accommodation will be made. The RTI program manager will be responsible for reconciling the AQS emissions data sets with the DQOs.

The emissions test results will be reported as  $mg/m^3$  (or  $\mu g/m^3$ ) TVOC and VOCs by species, unless all parties (EPA, RTI and vendor) have agreed that only TVOC shall be reported.

Appendix C: Method for the Evaluation of Emissions from Gypsum Board Using Environmental Chambers (AQS Method)

Available upon request.