The Advanced Animal Habitat-Centrifuge (AAH-C)

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ABSTRACT

The Advanced Animal Habitat - Centrifuge (AAH-C), currently under development by STAR Enterprises and SHOT, Inc., for the NASA Ames Research Center, is a controlled-environment accommodation for microgravity biological research with rats or mice and, potentially, other rodents. The AAH-C is designed primarily for long-duration operation on the International Space Station (ISS) and also for operation in the Shuttle Middeck and SPACEHAB™. The AAH-C is transported to and from the ISS in Avionics Bay 3A of the Shuttle Modified Middeck (with biospecimens), or in the Mini-Pressurized Logistics Module (without biospecimens). The AAH-C, which is modularized and internally reconfigurable, is capable of accommodating mice and rats from weanlings to adults. The Habitat is designed for operation in the ISS Habitat Holding Rack (HHR) (providing a microgravity environment), on the 2.5m Centrifuge Rotor (providing a selectable acceleration environment from microgravity to 2.0g), and at the Life Sciences Glovebox (providing for experimental procedures and unique specimen manipulations). The AAH-C provides all of the systems (food and water delivery, waste management, lighting, video, and environmental control) necessary to maintain optimal animal husbandry, while maximizing scientific output. The AAH-C is currently scheduled for Increment 17A launch in October, 2005.

INTRODUCTION

The study of laboratory animals is a long-standing goal of research aboard the International Space Station (ISS). Opportunities to observe rodent behavior, physiological variables, neurovestibular effects [Ross, 1993], bone and muscle loss [Backup et al., 1994], immune status [Lesnyak et al., 1996], blood chemistry and functional development [Walton et al., 1997] in low gravity are unique to the ISS. Experience on short missions of the Space Shuttle using NASA Animal Enclosure Modules (AEMs) [Walton et al., 1997], longer-duration Bion-series missions [Ilyin, 2000] and animal exposures to ground-based gravitational facilities [Bikle et al., 1994] clearly point the way to the need and procedures for longer-duration continuous studies of laboratory rodents on orbit.

The Advanced Animal Habitat - Centrifuge (AAH-C) is currently under development by STAR Enterprises and SHOT, Inc., for the NASA Ames Research Center as a component of the Habitat Holding Rack (HHR), a science double rack that will also accommodate the International Space Station's Cell Culture Unit (CCU), Plant Research Unit (PRU), Aquatic Animal Facility, Insect Habitat and Egg Incubator, collectively known as the Space Station biological Research Project [Johnson et al.. 1997]. The Habitat is designed for operation in the HHR providing a microgravity environment, on the 2.5m Centrifuge Rotor providing a selectable acceleration environment from microgravity to 2.0g, and at the Life Sciences Glovebox providing for experimental procedures and unique specimen manipulations.

AAH SYSTEM OVERVIEW

General Features - The AAH-C is a controlled-environment accommodation for microgravity biological research with rats or mice and, potentially, other rodents. The AAH-C is designed primarily for long-duration operation on the International Space Station (ISS) and also for operation in the Shuttle Middeck and SPACEHAB™. The AAH-C is transported to and from the ISS in Avionics Bay 3A of the Shuttle Modified Middeck (with biospecimens), or in the Mini-Pressurized Logistics Module (without biospecimens). The AAH-C, which is modularized and internally reconfigurable, is capable of accommodating mice and rats from weanlings to adults. The AAH-C provides all of the systems (housing and cage, food and water delivery, waste management, lighting, video, and environmental control) necessary to maintain optimal animal husbandry, while maximizing scientific output.

The AAH-C is of modular design, which provides for simple maintenance operations and facilitates the incorporation of scarring for future upgrades and performance enhancements. Specific AAH-C upgrades and enhancements planned include the following.

- Waste management system performance enhancement, from 20-days between changeout to 30-days between changeout
- Introduction of an animal biotelemetry system
- Introduction of individual housing and development inserts for rats and mice
- Introduction of a food pellet dispenser, in addition to the existing food bar and food plate dispensers
- Introduction of means of measuring quantities of food and water dispensed and consumed by the animals

Animal Health and Wellbeing Considerations – The AAH-C is an autonomous transportation and orbital life support system and laboratory, which must be able to support live rats and mice in a healthy environment that provides for their well-being. As such, the AAH-C design and performance must meet the criteria for the Guide for the Care and Use of Laboratory Animals (Institute of Laboratory Animal Resources Commission on Life Sciences, National Research Council, 1996). Further, expected variances with respect to established guidelines and practices, which are driven by the unique environment and constraints of the ISS, required approval by the Institutional Animal Car and Use Committee (IACUC). The AAH-C is the home of the animals, and it is necessary to provide them with a comfortable environment, which in turn will be conducive to the conduct of meaningful science.

AAH-C Design and Environmental Provisions - The AAH-C animal enclosure height and volume are in excess of 18 cm and 17,000 cm^3, respectively. Each AAH-C will accommodate up to six rats (weighing up to 600 grams each) or up to twelve mice (weighing up to 60 grams each) in group housing configurations, and up to two rats or two mice in individually housed configurations. The AAH-C provides programmable day and night cycles, to meet the anticipated range of animal circadian rhythm requirements, and incorporates both "white" and infrared lighting to enable light and dark cycle video monitoring of the animals for both science and animal wellbeing observations.

Air temperature in the AAH-C animal enclosure (or cage) is controlled within the range of 20°C to 30°C, and relative humidity is maintained to between 40% and 70%. The airflow within the animal enclosure is maintained between 0.1 to 0.3 meters/second, and the air pressure
is maintained at 50 to 75 Pa below cabin pressure. CO_2 and NH_3 concentrations are maintained at less than 1000 ppm and 25 ppm, respectively. Eight AAH-Cs will be available on-orbit, four hosted in the Centrifuge and four in the Habitat Holding Rack. See Figure 1.

Figure 1. Exterior design of the AAH – C. The main chassis is compatible with all space vehicles. For use on ISS the Chassis Extension Unit (CEU) is attached at the rear of the chassis and is used to store supplies and to effect heat transfer to avionics air.

Chassis – The AAH-C outer housing, or chassis is the structural backbone for the habitat, which provides a level of containment for the habitat subsystems and physical tie points for all internal and external interfaces. On the ISS, interfaces with the Habitat Holding Rack (HHR), Centrifuge Rotor (CR), and Life Sciences Glovebox (LSG) are facilitated by hand-operated fasteners.

Chassis Extension Unit (CEU) – For ISS Extended Duration Operations, the CEU attaches to the rear of the Habitat chassis and provides an extra volume for additional systems, including water reservoirs and biotelemetry electronics modules. The CEU is not utilized in Middeck locker and SPACEHAB™ configurations due to a limited locker depth in the x-dimension. The CEU is shown behind the Habitat in Figure 1.

AAH SYSTEM DESIGN

Cage System – There are two basic versions of the AAH-C internal Cage System design; one version is configured to support group-housed rats (bar spacing 0.5", floor grid 2 squares/inch), while the other is configured to support group-housed mice (bar spacing 0.33", floor grid 3 squares/inch). Each version is re-configurable, with the addition of appropriate inserts, to provide for individual and animal development housing. The Cage System incorporates access doors at the "top", which are opened when the AAH-C is attached to the (bottom of) the Life Science Glovebox (LSG). This enables animals to be removed for science manipulations, animal health and wellbeing inspections and Cage maintenance operations. In order to minimize the demands on the crew in maintaining a clean environment for the animals, the Cage is designed as a collapsible (foldable) unit. A used ("dirty") Cage is removed, collapsed and stowed, while the AAH-C is attached to the LSG, and a new ("clean") Cage is assembled and installed (no on-orbit cleaning time required).

The Cage System is subject to – and designed to function in – two different (orthogonal) g-vector orientations, one for transport in the Shuttle Middeck and the other for on-orbit operations. When mounted in the Shuttle Middeck at launch, the g-vector is oriented towards the rear of the Cage, which thus becomes the "floor" for the animals. In this (transport) configuration, the Cage has two transition filters installed for waste management, one at the rear and one midway between the front and rear of the Cage, forming two "compartments" for the animals. By this means, the effective floor area is doubled in the transport configuration, thereby providing for accommodation of twice the on-orbit complement of animals. Each transition filter comprises an "upper" surface grid, which supports the animals, and a multi-layered waste filter, separated from the grid by an air gap that prevents direct animal contact with their feces and urine. Once on orbit, the transition filters are removed (during the first animal access operation) for bagging and stowing with the launch cage.

The AAH-C Cage System, when configured to support group-housed rats or mice, accommodates a maximum number of animals, which depends on the collective biomass and the configuration (on-orbit or transport), as follows:

Specimens	Specimens Supported (Per Habitat)	
	Nominal (On-Orbit)	Transport (Middeck)
Mice (3-14 gm)	12	-
Mice (16-50 gm)	12	18
Rats (30-100 gm)	6	16
Rats (101-400 gm)	6	12
Rats (401-600 gm)	4	8

Each Cage configuration complies with (in fact, exceeds) the following sizing requirements:

- Minimum cage height: 18 cm (actual height: 20.32 cm)
- At least one dimension (e.g floor diagonal): 21 cm (actual floor diagonal: 43.9 cm)
- Minimum cage volume: 16,546 cm^3 (actual cage volume: 18,533 cm^3)

Features of the Cage System design (rat grouped configuration) are shown in Figure 2.

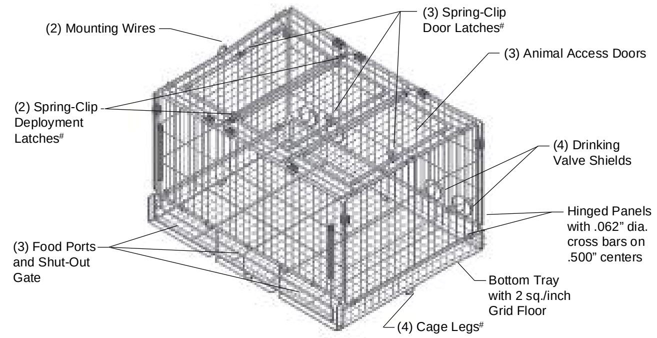

Figure 2. One of several internal cage configurations for AAH – C. Dimensions satisfy animal-care requirements. Access doors provide access to Life Sciences Glovebox on ISS.

Food Delivery System – The Food Delivery System consists of two interchangeable assemblies, a Food Bar Dispenser and a Food Bar Plate, both of which provide on-demand solid nourishment to the rodents while minimizing wastage. The Food Bar Dispenser provides food to the animals for 11.5 days (design goal); whereas the Food Bar Plate provides food for 14 days.

The Food Bar Dispenser assembly consists of the following:

- A light-weight housing for the foodbars
- A (food bar) pusher sub-assembly, powered by a constant-force spring
- Ramps (which, together with cut-outs on either side of the food bar, prevent "pillaring")
- Tunneling pins (which prevent animal tunneling into the food bar)
- A sweep mechanism (which moves animals out of the feeder alcove before Cage removal of food module changeout)

The Food Bar Dispenser is equipped with a closeout plate that provides the ability to close off food ports during module change out. The Food Delivery System configuration facilitates change out/refill from the front of the habitat (without use of a glovebox). The Food Bar dispenser assembly is illustrated in Figure 3. Figure 4 shows the food bar shape.

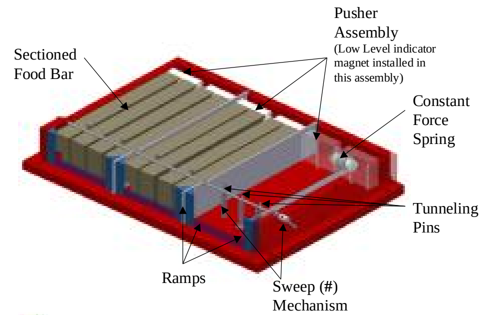

Figure 3. The Food Bar Dispenser assembly has the capacity to provide the animals with on-demand food for almost 12 days.

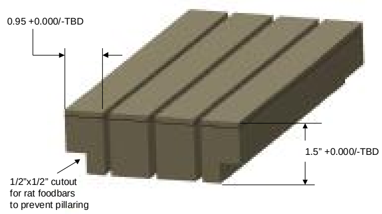

Figure 4. The Food Bar configuration incorporates a cut-out to prevent "pillaring".

The Food Bar Plate assembly consists of a stainless steel plate (to which the food bar is glued), a locking mechanism (for assembly attachment to the cage) and grasp rings (for the animals to grip when feeding). The food bar plate assembly is illustrated in Figure 5.

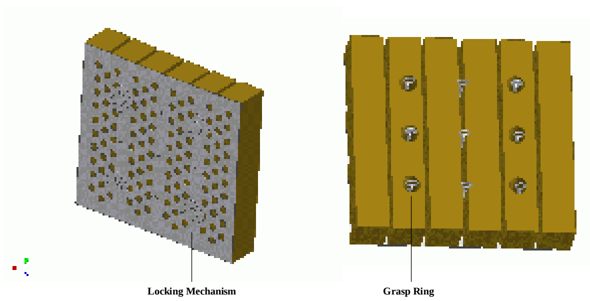

Figure 5. The food bar plate assembly has the capacity to provide the animals with on-demand food for 14 days.

Water Delivery System – The Water Supply System dispenses potable water "on demand" via lixits fed from pressurized reservoirs. The system consists of the following:

- Five reservoirs (one in the habitat, four in the CEU)
- Cross-plumbed supply lines (for redundancy) – PFA (Teflon) tubing and fittings
- Iodinators (4 ppm in-line and 10 ppm fill) to iodinate water and reduce bacteria counts
- Flow sensors
- Valves (isolation, manual shut-off, check)
- Main/CEU accumulator and water delivery manifold assemblies
- Front panel and CEU Quick Disconnects (QDs)

The main/CEU accumulator assemblies have a total capacity of 1.25 liters of usable water, and consist of stainless steel bellows accumulators, each incorporating string potentiometers, which provide water quantity measurement. The accumulator pressure range is from 10 psig (maximum), when full, to 2 psig (minimum), when almost empty.

The water delivery manifold assembly consists of four mini-accumulators (bellows type), a lightweight plastic manifold, four SE Labgroup ML-125 drinking valves, and four latching solenoids (one per mini-accumulator). The valves open, allowing water to flow from the main to the mini-accumulators. The solenoid valves close once the mini-accumulators are full, isolating the drinking valve and mini-accumulators from the main accumulators. The mini-accumulators provide 0.5 to 1.0 psig pressure at the drinking valves. A string potentiometer is attached to each mini-accumulator to provide leak detection, measure water quantity and control automatic refill.

Quick disconnects attach the Habitat water lines to the CEU reservoir. Lixits allow animals to orally apply a small pressure drop across the porous terminus of the water lines to obtain drinking water. Habitat and CEU water reservoirs are refilled on orbit from the front of the Habitat.
The capacity of the water delivery system is sufficient to provide the animals with water for 15 days on the ISS.

The Water Delivery System assemblies are illustrated in Figure 6. Figure 7 shows the Water Delivery System as installed in the AAH-C.

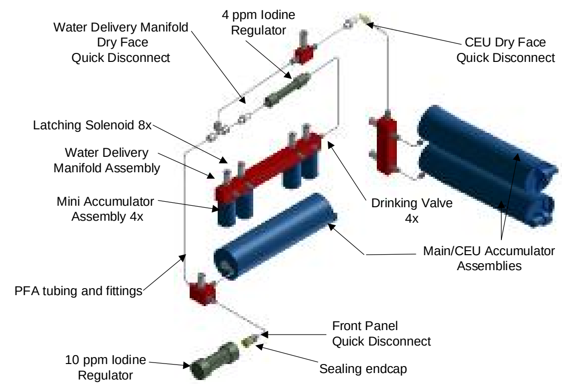

Figure 6. The AAH-C Water Delivery System provides "on-demand" potable water to the animals, fed from pressurized accumulator assemblies.

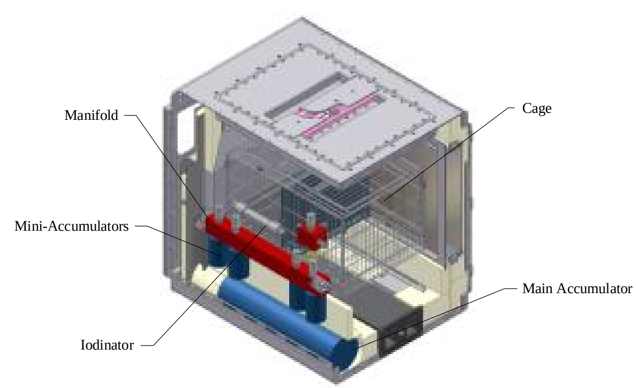

Figure 7. The AAH-C Water Delivery System installed in the Habitat is configured to provide for water replenishment at the front panel

Waste Management System – The primary function of the Waste Management System is to collect, contain and process urine, feces, dander, loose fur, glandular secretions, food particles and dust, and water (potable and condensate). The Waste Management System incorporates three unique types of filter:

- A waste filter for collection/containment of all solids and liquids
- Bleed air and specimen chamber inlet filters, and a bleed air exhaust filter for containment of animal, food-bar and waste odors
- A transition filter for temporary waste management during transportation in the Shuttle Middeck.

The main waste filter separates liquid and solid waste, and rapidly evaporates and removes liquid in the bleed air exhaust. There are three distinct sections in the main waste filter: a solid waste screen, which captures solid waste and prevents clogging of the liquid separator/evaporator passages; a liquid separation and evaporation section, which collects waste water and facilitates removal by evaporation; and a pleated particulate post-filter, which removes 90% of particles larger than 25 microns.

The bleed air inlet filter removes 97% of all particles larger that 0.3 microns. It is of pleated design to minimize the pressure drop and maximize the surface area. It is replaceable from the Habitat front panel. The specimen chamber inlet filter removes 90% of all particles larger than 3 microns. It is also of pleated design and incorporates a urine deflection screen installed to protect the filter media. The filter is removed and replaced between mission increments. The bleed air exhaust filter is of multi-layered construction. The outer layer removes 97% of particles larger than 0.3 microns; the intermediate and inner layers inhibit the passage of ammonia, animal waste and food bar odors from inside the AAH-C to the crew cabin.

The specimen chamber liner covers the specimen walls from the lid to the top of the waste filter to minimize fouling of the interia habitat housing.

Waste Management System design features include protection of animals from harmful volatiles produced from decomposing waste, prevention of malodorous, harmful, gaseous and particulate materials from entering the crew environment, waste collection in two different g-orientations (with and without gravity) and prevention of animal access to waste. A functional block diagram of the Waste Management System is shown in Figure 8. Figure 9 shows a view of the AAH-C with the Waste Management filters in place.

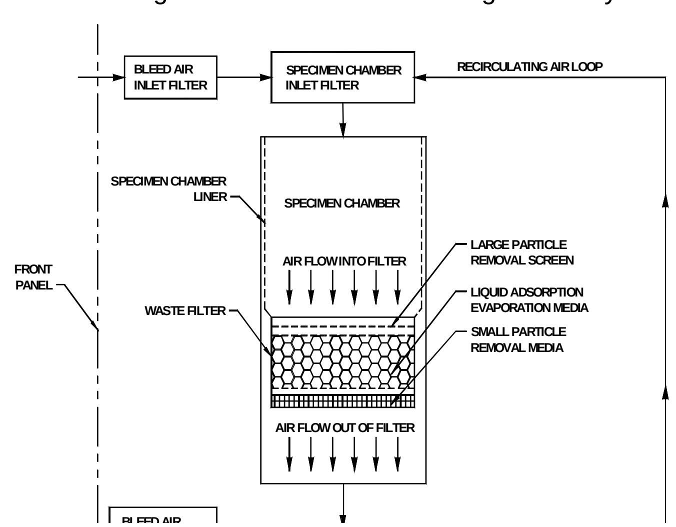

Figure 8. Functional block diagram of the Waste Management System (on-orbit configuration). In the on-orbit configuration, the system incorporates bleed air inlet and exhaust filters, a specimen chamber inlet filter, and a waste filter.

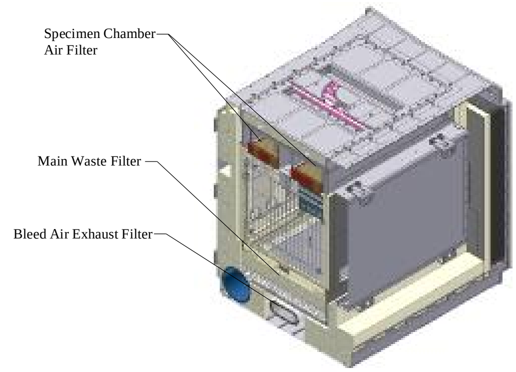

Figure 9. The AAH-C Waste Management System installed in the Habitat provides an effective mechanism for the collection and processing of solid and liquid waste, and the containment of odors released to the cabin.

<u>Environmental Control System (ECS)</u> – The ECS exercises control or maintenance of the specimen chamber environmental conditions, such as air flow rate, air temperature, cabin air exchange, and relative humidity. Specifically, the ECS performs the following functions:

- CO_2 maintenance
- Relative humidity maintenance
- Specimen chamber heat rejection and temperature control
- Specimen chamber air flow and pressure drop maintenance
- Avionics volume air flow and heat rejection
- Condensation containment

Optimization of the thermal environment is achieved with three independent thermal control loops: an electronics thermal loop, a heat exchanger thermal loop, and an animal enclosure thermal loop. Air temperature in the AAH-C animal enclosure is controlled within the range of 20°C to 30°C, in 1°C set-point encrements. The temperature control requirements levied on the animal enclosure temperature control system are the following:

- Accuracy: ±1°C, with respect to the set point
- Isothermality (empty animal enclosure): ±2°C
- Maximum overshoot/undershoot during set point transitions: 2°C

Closed-loop control is exercised by passing fan-driven air over Thermal Electric Devices (TEDs) for heating/cooling (air-to-air heat exchangers). Multiple temperature sensors (for redundancy) provide the temperature feedback signals for the closed-loop control. The variable speed fans circulate air through both sides of the heat exchanger and provide cabin air exchange.

Relative humidity sensors provide measures of the levels at key locations within the AAH-C. Relative humidity is maintained between 40% and 70% by means of a software override of the temperature set point, which increases the set point temperature to lower the humidity whenever it exceeds 70%, and decreases the set point temperature to raise the humidity whenever it falls below 40%. For example, an increase in temperature set point from 20°C to 25.5°C will drop the relative humidity levels from 98% to 70%. The variable speed fans provide airflow to facilitate evaporation and provide cabin air exchange to reject water vapor to the cabin. The airflow within the AAH-C animal enclosure is maintained to between 0.1 to 0.3 meters/second and the air pressure is maintained at ≥ 50 Pa (0.2 inches of water, 0.007 psi) below cabin pressure, which provides a level of containment of the contents of the animal enclosure. CO_2 and NH_3 concentrations are maintained at less than 1000 ppm and 25 ppm, respectively. A three-dimensional model projection, illustrating the ECS design architecture, is shown in Figure 10.

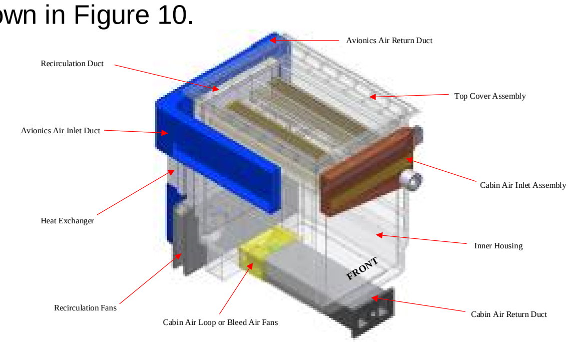

Figure 10. The AAH-C ECS exercises authority over the temperature, relative humidity, pressure and quality (constituents) of the air flowing within and through the Habitat

Illumination and Video - The AAH-C provides programmable day and night cycles, to meet the anticipated range of animal circadian rhythm requirements, and incorporates both "white" and infrared lighting to enable light and dark cycle video monitoring of the animals for both science and animal health and wellbeing observations. The Illumination System enables control of the light level in the specimen chamber between 0 and 40 lux, with at least four set points. It provides uniform light (±20%) to the entire cage to monitor animal activity/behavior during day and night cycles. Both "white-light" and infrared LEDs (for dark-cycle monitoring) light the cage area utilizing surface mount technology. The spectral irradiance of the white-light LEDs is distributed with 50% continuously distributed between 400 and 580 nm and 20% continuously distributed between 580 and 700 nm.

In addition to the 0 to 40 lux capability, the Illumination System incorporates provision for the operator to raise the light level throughout the animal enclosure to 100 lux, for short periods of time, to facilitate direct visual checks of the health and wellbeing of the animals.

The Illumination System design utilizes two linear lighting modules, approximately 1.6 cm wide, and 33 cm long. The modules are mounted on polycarbonate diffusers on top of the AAH-C lid assembly, and are populated with Light-Emitting Diodes (LEDs) and photo sensors. The selected illuminance is generated by Pulse-Width Modulation (PWM)

The Video Monitoring System consists of three board cameras centered above the Cage. The cameras are a combination of black and white and color, and are vertically-mounted in the AAH-C lid. The Video Monitoring System provides both day and night imaging by utilizing the white-light and infrared illumination systems, respectively. Since the cameras' viewing windows are expected to foul, a scrolling mechanism, which provides a continuous clear mylar viewing surface, is included in the design. By using remote electronics the thermal load due to video operations is minimized within the specimen chamber. Video coverage includes at least 90% of cage volume.

The Illumination and Video System configuration is illustrated in Figure 11.

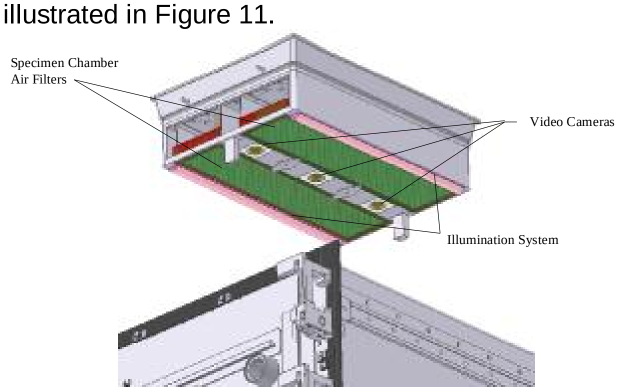

Figure 11. The Illumination and Video System, installed on the top of the Habitat lid assembly, provides uniform lighting and in excess of 90% video coverage of the animal enclosure internal volume

SCIENCE EVALUATION UNIT

At the current state of development a ground-based equivalent AAH-C ("Science Evaluation Unit" – SEU) is under construction, and it will be used in tests of all subsystems using the appropriate numbers of mice and the appropriate numbers of rats as test objects. The SEU approaches being a high-fidelity replica of the final AAH-C design and is certainly suitable for use as a ground-control habitat. It should also be noted that the SEU habitat is designed for the approved maintenance of mice and rats, totally unattended, for time periods as long as a few weeks. Thus the SEU is a useful tool in experiments in which undisturbed animal maintenance is required, yet NIH animal-care guidelines must be met in non-space applications. A rendering is shown in

Figure 12.

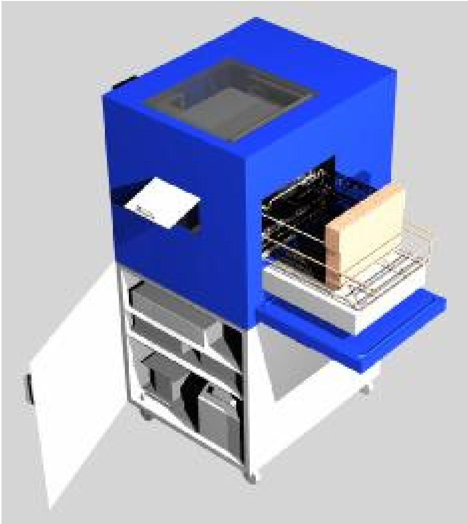

Figure 12. Advanced Animal Habitat Science Evaluation Unit (SEU). Rendering of the exterior of the current design illustrates ground-based configuration that will fit on a movable stand (top) and attach via interface cable to the command-and-data module on a separate stand (not shown).

DEVELOPMENT TESTING

Several AAH-C subsystem and system prototypes have been developed and tested during the AAH-C preliminary design phase of the development program. Emphasis has been placed on biospecimen testing to evaluate the evolving designs, particularly with respect to their functionality and compliance with the animal health and wellbeing requirements. Tests conducted on development units and prototypes, with animals, include the following:

- Prototype cage prototype designs - observations and evaluations of animal adaptation, behavior and health
- Water Delivery System – observations and monitoring of animal use and water consumption
- Food Delivery System – observations and monitoring of animal use and food consumption, with particular attention focused on any tendencies for the food bar to jam, "pillar" and form alcoves into which the animals could "tunnel", or become trapped and/or injured.
- Waste Management System – evaluation of effectiveness and performance in collecting and processing animal waste and containing odors
- Environmental Control System – evaluation of effectiveness and performance in controlling the temperature within the animal enclosure, maintaining the relative humidity levels within the specified range, maintaining the required airflow through the animal enclosure, and containing the levels of carbon dioxide ammonia and malodorous constituents within and released from the cage.

Development testing, particularly involving animals, is vital in the process of establishing a Habitat design that is fully compliant with system configuration and performance requirements and optimally responsive to the established animal health and wellbeing requirements.

CONCLUSION

The AAH-C provides a complete biological research environment for rodents for long-duration operation on the International Space Station (ISS). It is internally modularized and reconfigurable and is capable of accommodating mice or rats from weanlings to adults.

The modular Food Delivery system provides on-demand solid nourishment (foodbars) to the animals while minimizing wastage, and the Water Delivery System dispenses potable water on demand via lixits fed from pressurized reservoirs.

The Waste Management System collects, contains and processes urine, feces, dander, loose fur, glandular secretions, food particles, water (potable and condensate). It also prevents animal access to waste products and protects them from harmful volatiles produced from decomposing waste. It further prevents malodorous, harmful gaseous and particulate materials from entering the crew environment

The Lighting System provides control of light levels in the specimen chamber, and is programmable to provide day and night cycles. The Video monitoring system provides both day and night high-resolution imaging for animal viewing.

The Environmental Control System provides control of air flow, and temperature, and the regulation of relative humidity within the Habitat. The system also provides for the monitoring and regulation of ammonia and carbon dioxide within the Habitat and contains the levels of airborne malodorous contaminants released from the Habitat.

In developing the AAH-C design full advantage has been taken of the latest applicable research and technology, and the Habitat, as such, offers a significant advancement relative to the previous generation of animal habitats.

ACKNOWLEDGMENTS

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DEFINITIONS, ACRONYMS, ABBREVIATIONS

AAA	Avionics Air Assembly
AAH-C	Advanced Animal Habitat – Centrifuge
CEU	Chassis Extension Unit
CCU	Cell Culture Unit
CO_2	Carbon dioxide
HHR	Habitat Holding Rack
ISS	International Space Station
LED	Light-emitting diode
NH_3	Ammonia
NIH	National Institutes of Health
Pa	Pascals of pressure (Newtons/m^2)
PRU	Plant Research Unit
SEU	Science Evaluation Unit
SHOT	Space Hardware Optimization Technology, Inc.