Infrastructure Choices and Their Relationship To Capital Equipment, and Basic Operation of a Zebrafish Facility

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Today's zebrafish facilities scarcely resemble the Quonset huts of the Streisinger Era or the dank basements common to older university aquatics labs. Indeed, modern zebrafish labs are rife with cutting edge technology, from automated feeding systems and chemical cage wash, to light-sheet microscopes and complex behavioral research suites. When building a new zebrafish facility, either in a renovated space or in an entirely new building, the choices you make regarding the various infrastructure components will have a significant influence on the outcome of your project.

Structural Considerations: While not strictly an infrastructure component, one of the engineering considerations that must be accounted for before construction is the substantial weight of the equipment and the water it carries. When designing the floors, walls, and ceilings, it is critical to consider the overall weight and distribution of the water in fish housing and life-support rooms, and the local requirements for seismic restraints on such equipment. These load capacities should be exceeded where possible to accommodate any future expansion of the operations.

HVAC: While there is not a great deal of consensus about air temperature, humidity, and air exchange rate in zebrafish facilities, it is fair to say that we are past the era of hot and sticky fish housing rooms. With the HVAC technology available today, it is entirely possible to keep the work areas comfortable for the people, while allowing for the complex recirculating aquaculture systems (RAS) to maintain perfect conditions for the fish. Ambient temperatures in fish housing and procedure rooms should range from 23-26C depending on the needs of the research, and relative humidity should be kept at or below 50% where possible, and not exceed 70%. And while inadequate air exchanges foster accumulation of moisture which supports fungal growth, excessive air exchanges increase evaporative losses from the fish systems increasing their operating costs, therefore air exchanges should be maintained at approximately 6-9 ACH to avoid these extremes. Whenever possible, HVAC systems should have automation and BMS incorporated to ensure that facility managers can be alerted of deviations from the normal operating ranges which can quickly create animal welfare concerns.

Electrical Supply: The voltage and amperage of the electrical supply will be dictated by the utility requirements of the capital equipment you plan to purchase, along with the sum total of the anticipated ancillary equipment and lighting needs., and Tthe facility manager will have critical information to contribute in this area- so be certain to ask them. Careful attention should be paid to the specifications of the capital equipment as it often varies significantly from one vendor to the next, and may be manufactured in a country where voltages are different from that of your region. It is preferable to have higher voltages supplied to the space and then employ transformers to reduce voltages, rather than the other way around, due largely to the escalation in amperage resulting from transformation in this direction. Sensitive equipment may require UPS and power conditioning to keep from invalidating warranty's warranties, and the presence of water and high humidity will dictate that non-metallic conduit and fittings should be specified. Emergency power should be supplied to all aspects of life-support systems and water purification systems, as well as ultra-cold freezers, incubators, and food production and storage areas. The sudden transition from normal power to emergency power can be rough and may also need to be accounted for when working with your equipment vendors.

Plumbing: Plumbing is best approached by separating it into two distinct camps: water supply, and water drainage. The municipal or mains water supply in zebrafish facilities must further be separated into two distinct groups: direct use, and pre-purification source water.

The direct use water may be best considered as that which you will find at typical hand-washing sinks and restrooms. Little if any further filtration or purification is required with this water.

The pre-purification source water is best considered as the water that must undergo considerable purification before it can be used to fill the recirculating fish systems, autoclaves, and automated cage-wash equipment. The acceptable level of purity of most municipal drinking water, in combination with the chemicals used to make that water safe for us to drink, is toxic to most fish, including zebrafish. As a consequence, most zebrafish facilities opt to purify this water using reverse osmosis (RO), and de-ionization (DI), equipment, and we often design in redundancy from the start. The RO/DI equipment will have several requirements of the water that enters it, often resulting in several steps of pre-treatment, as well as specific electrical and data needs. Usually starting with tempering, additional pre-treatment steps often include sediment filtration, softening, and carbon filtration. RO/DI water is both slow to produce and costly to make, and this will necessitate substantial storage of purified water to meet the daily needs of the facility at full operating capacity, while always considering the emergency scenario where the purification system is down for a period of time. It is important to understand that RO/DI is very corrosive, and will require special material choices for all piping, fittings, and fixtures with in which it makes contact. There are only a couple of options for the plastic pipe which are compatible with RO/DI water, and reservoirs and recirculation pumps should also be specified which meet these same compatibilities. A failure to do so can result in water that is polluted with a contaminant, and catastrophic failure of plumbing components.

The plumbing concerned with drainage in zebrafish facilities is an area where planning for the worst-case scenario is your best bet. In aquatics laboratories it is not a question of *if* there will be a large flood, but *when* it will happen. Trench or trough style drains that cover the entire width or length of a fish housing room are the superior choice, rather than placing a few circular floor drains around the room. This not only allows for larger spills or leaks to be efficiently contained, but also allow for more consistent and uniform slopes to be applied to the floors during construction. These trench drains should be made from fiberglass or plastic, and be covered with either 316-stainless steel or non-metallic covers to prevent corrosion. Other areas where the materials choice, location, overall size, and capacity of the drains are important to consider are in the water purification area, where the RO/DI equipment will have a high rate of discharge, in the mechanical areas where the life-support systems are that maintain the zebrafish colony, and in the cage-wash area, where all of this specialized equipment will come with specific demands. In areas where large amounts of water are regularly drained, such as live-feeds production areas and cage-wash staging areas, pit-style drains with plastic grates are a great choice, but they must be integrated into the design early in the process.

Steam: While not every building has the option to supply steam, when it is available, it can be a great resource for some of the specialized equipment found in zebrafish facilities. In addition to simplifying the specialized HVAC demands, steam is the superior choice for heating in autoclaves and cage-wash equipment. However, it is important to understand that there are different grades of steam available, and depending on your needs, you may need to make adjustments to the supply you have. Autoclaves and cage-wash machines will require “filtered” or “clean” steam that is free of amines and cleaning and conditioning chemicals, to prevent carryover to the fish systems. And the various valves and fittings in this equipment need to be protected from solid debris in the steam lines by the addition of particulate filters. Failing to comply with these guidelines may result in mechanical failures and serious health consequences for the fish.

IT: Our world today is increasingly connected, and zebrafish facilities are no exception. In addition to the normal computing needs, virtually all of the capital equipment and much of the microscopes and other procedure-oriented equipment have wireless or Ethernet capabilities. These complex pieces of technology can often be managed and programmed remotely, and the network infrastructure needs to be designed to maximize this advantage while protecting any sensitive data that may be contained in the institution. Colony management databases often use wireless devices, including phones and tablets, to communicate with software running on a computer in the facility. When you consider the vast amount of water, concrete, and metal that connects and surrounds a modern zebrafish facility, you will quickly realize that creating a strong wireless coverage area is a challenge, and may require a high degree of redundancy to ensure good signal strength.

Compressed Air: Compressed air is an invaluable, but often overlooked resource in a zebrafish facility. With the addition of regulators and filters in the proper locations, it can be used to provide supplemental aeration to life-support systems and maintain live feed culture operations. It can also fully replace the compressed nitrogen tanks (N_2) often used to supply gas pressure to the pico-pumps at micro-injection stations., and And compressed air is often required for cage-wash and autoclave machines where the equipment relies on it to perform many aspects of its operations. The pipe used to carry compressed air should be suitable for a wet-environmentwet environment, this means no copper or galvanized steel, and the filters should be designed to render clean (oil-free) and dry air.

Natural Gas: In addition to typical HVAC needs, many zebrafish labs still contain procedure rooms, bench science areas, fume hoods, and bio-safety cabinets where natural gas is still used for burners and it will need to be available for the lab valves common to this type of equipment. Natural gas may also be needed at the increasingly common tank-less water heaters found in areas where routing hot water may be inefficient.

Liquid Nitrogen (LN_2): Zebrafish sperm is often archived using cryo-preservation techniques where liquid nitrogen is employed to maintain the very low temperatures needed. Doing so provides a contingency plan should a fish of specific genetic background or particular interest be lost to an unplanned event, and may also be a component of the overall genetic management of stocks within a colony. In some cases LN2 can be piped directly to filling stations and storage tanks which contain the samples, but in many cases large cylinders containing the LN2 will need to be delivered, connected to the storage tanks, and replaced when depleted. This creates a situation where regular involvement with the equipment is required by the staff. One way to streamline this otherwise arduous task is to provide a docking station and manifold to connect the cylinders to the storage tanks.