BIOREGENERATIVE FOOD PRODUCTION SYSTEM: USING INTEGRATED FOOD PRODUCTION SYSTEMS TO FEED THE FUTURE.

by

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Nomenclature

AF = Animal Feces

AR = Area Required

AW = Animal Waste

BE = Bio-efficiency

BFPS = Bioregenerative Food Production System

BLSS = Bioregenerative Life Support System

BVAD = Baseline Values and Assumption Document

CEA = Controlled Environment Agriculture

DW = Dry Weight

ER = Energy Required

FCR = Feed Conversion Ratio

FF = Fish Food

FW = Fresh Weight

HI = Harvest Index

IPM = Inedible Plant Material

MW = Mealworm

NW = Net Waste

OM = Oyster Mushroom

SMS = Spent Mushroom Substrate

TAN = Total Ammonia Nitrogen

TN = Tilapia

TSS = Total Suspended Solids

TW = Total Weight

WC = Water Content

WR = Water Required

WW = Wet Weight

Abstract

The challenge of feeding a growing population requires increased production efficiency, especially when production is extended into suboptimal production environments. In such areas, resource conservation is increasingly important and innovative linkages between waste streams and food streams can significantly impact total system efficiency. Bioregenerative Life Support Systems (BLSS) for space have been designed in which plants and mushrooms are grown in tandem, maximizing total food produced, minimizing inedible waste, and maximizing energy and space efficiency. The purpose of this work was to evaluate application of BLSS for food production on Earth as Bioregenerative Food Production Systems (BFPS) For this project, a model was created to predict productivity of a BFPS with production components for plants, mushrooms, insects, and fish, while feeding a single person for one day, and a specific population for a specific set of time, according to the NASA BVAD document. The waste outputs of one production module become the inputs for the subsequent module, ideally creating a closed nutrient cycle. Input for this model relied on data from primary literature, government agencies, and industry websites.

Input data was accumulated in a cornerstone data page and all component production pages referenced this cornerstone page for calculations. The first production page included only plant components; subsequent components were added onto prior production runs in an iterative process. Each production run was compared to previous and subsequent production runs for water, energy, and growing space efficiencies, and net waste products.

Chapter 1: Introduction to BFPS

Food Crisis

The world's population is approximately 7.8 billion [1], and that number is predicted to grow to 9.7 billion by 2050 [2]. Feeding this growing population continues to be a challenge, particularly in terms of land use [3]. Currently, approximately 50% of the total arable land on earth is used for agricultural purposes, with livestock accounting for 77% of agricultural land use [3]. However, the Food and Agricultural Organization of the United Nations (FAO) forecasts that food production must increase by 70% by 2050 to meet the nutritional needs of the population [4]. As it stands, there is simply not enough arable land to feed the predicted population increase with conventional farming methods without exacerbating environmental damage and climate change. New methods of Agriculture are needed.

New agricultural technologies: Controlled Environment Agriculture

To produce more food predicably on a smaller physical footprint, modern agriculture relies on high-yielding practices while reducing production variables. This is increasingly accomplished using controlled environment agriculture (CEA) [5]. CEA is focused on controlling all environment and environmental inputs of a defined growing space such as a greenhouse or an enclosed vertical farm. CEA allows for the optimization of plant growth through the manipulation of production inputs, and the ability to grow crops virtually anywhere year-round. Moreover, CEA systems placed in urban centers can reduce the need for transportation and subsequent costs in time and fuel.

Hydroponics

Modern greenhouse production is often accomplished without soil via hydroponic technology, which is the cultivation of plants using nutrient- and oxygen-rich water instead of soil [6]. Often a root-supporting substrate is used to support the plants as they grow. However, some systems provide no support at all and plants simply floating on a nutrient solution (deep-bed culture [7]). Other forms of hydroponics include Eb-and-flood systems and nutrient film technique systems (NFT).

Vertical Farming

Efficiencies of such systems are maximized when production unit are stacked vertically with supplemental lighting in completely enclosed controlled environments [8]. This format is termed vertical farming. Vertical farming allows for large amounts of growing space for crops while minimizing the actual area occupied by the growing facility, making such a system feasible for location within the limits of a city or town. Despite these technological improvements, plant-based CEA retains one drawback compared to traditional field agriculture: There is a significant amount of inedible plant material leftover as agricultural waste. This is where mushroom farming can be applicable.

CEA in Mushroom Production

Certain crops are already produced predominantly in CEA facilities and have been for decades. Most commercial mushroom production occurs in enclosed facilities in which the light, temperature, humidity, and CO2 are closely monitored and tightly controlled [9]. Commercial mushroom production is one of the most valuable specialty crops based upon annual sales and is one of the fastest growing specialty crop commodities. In 2015, commercial market analyses placed world production value at approximately $30 B annually. Coupled with a 10-15% projected annual growth, the industry is positioned to surpass $50-70 B by 2024 [10]. As decomposers, mushrooms can be grown on a variety of substrates, including agricultural wastes [9]. Like plant-based agriculture, however, there is a waste product. In the case of mushroom farming, it is the spent mushroom substrate leftover from harvesting the mushrooms. This is where insect farming can be applicable.

Aquaculture and Insect farming

In addition to plant and mushroom production, other areas of agriculture include aquaculture and insect agriculture. Aquaculture is the cultivation of edible fish in either enclosures in open water, or in manmade pools or raceways. Aquaculture is widely reported to be the fastest growing food production sector world-wide, and aquaponics is one of the fastest growing subsectors. Multiple use of producing fish in irrigation water and use of fish effluent to irrigate and fertilize crops in a sustainable manner has been a hallmark of our research and subsequent extension to arid and semi-arid regions around the world [11]. Edible insects are recognized by the FAO as an important source of protein for human consumption [12]. Insect farming, a food industry with exponential growth, is recognized as a priority by the USDA [13]. Insects are rich in protein and minerals and require much less water, land and feed than do vertebrate livestock. [3][14].

Aquaponics and Sustainable Animal Feeds

For enhanced efficiencies, systems can also be linked in CEA facilities. For example, aquaponics, the cultivation of fish or other marine lifeforms in conjunction with the cultivation of plants [15]. In aquaponics, the waste produced by the fish – largely ammonia - is converted by naturally-occurring bacteria in the system into nitrates which are usable by plants. The nitrate-rich water is then diverted to the plants, sustaining them while also cleaning the water. Aquaponics has several advantages over traditional field agriculture, such as a 90% reduction in water use, reduced use of fertilizers, eliminated risk of soil-borne diseases, and the ability to be grown in any location with access to electrical energy [15]. A second focal point has been the development of sustainable aquaculture feeds. Replacing fishmeal in aquafeeds is one of the most critical issues of aquaculture. Insects, fungi, algae, and composted plant materials are some of the key alternative ingredients that have been promoted as fishmeal substitutes and use of aquaculture effluents as nutrient rich organic fertilizer brings together the best parts of the Green and Blue Revolutions [16].

Closed Ecological Systems

An extension of CEA is the closed ecological system [17]. In a closed ecological system, an ecosystem is sustained in a controlled environment, with minimal exchange of gases and materials between the enclosed ecosystem and the external environment. One of the largest examples was Biosphere 2 in Oracle, Arizona, in the United States of America. This was an experiment attempting to sustain 8 people [18] in an enclosed, artificially maintained biosphere for an extended period, as a prototype for space colonization. The concept of sustaining humans using closed ecological systems has been explored several times since Biosphere 2. One such project developed at the University of Arizona is the Mars Lunar Greenhouse (MLGH), which was a greenhouse designed to provide both food and life support for humans on an extraterrestrial colony.

Introduction to the Bioregenerative Life Support System.

Gellenbeck et al. [19] explored the concept of a Bioregenerative Life Support System (BLSS) in 2019. This concept built off the MLGH's objective: to provide food and life support for human crews using biology-based technology. As a form of closed ecological system, the overall goal of a BLSS is to create a self-contained artificial ecosystem within a space habitat or vessel. Instead of simulating a natural ecosystem in a terrarium or similar structure, each component of the ecosystem is in a self-contained component. The outputs of one system become the inputs for another, effectively creating an artificial food chain or web with each component producing food that is edible to humans. In addition to food production, an ideal BLSS would consume sufficient carbon dioxide and produce enough oxygen to maintain a hypothetical human crew. The BLSS initially relied on plant production, which left a large amount of leftover inedible plant biomass. Gellenbeck's goal was to demonstrate the feasibility of adding an edible mushroom module to the BLSS as a means of converting the inedible biomass to edible mushrooms. He demonstrated that using waste plant material to produce mushrooms would lead to a 33% decrease in growing space and a 31% reduction in electrical consumption.

Project Scope: Bioregenerative Food Production System

The purpose of this project is to explore the use of a BLSS for terrestrial food production (now termed a Bioregenerative Food Production System (BFPS)). This project expands on the work done by Gellenbeck et al [19] with the inclusion of components for insect and fish production to further utilize system waste streams as resources for alternative food production. The insect component relies on feeding edible insects with the spent mushroom substrate leftover by the mushroom component. The insects are then combined with any uneaten substrate and fed to the fish. Leftover substrate is diverted to another component, potentially for heat and electrical energy production; leftover mealworms are diverted to human consumption. These additional components have the potential to further reduce growing space, energy consumption, water use, and waste production, while also maximizing food production.

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Chapter 2: Biological Food Production System Research and Modeling

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Bioregenerative Food Production System: Using integrated food production systems to feed the future.

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I. Abstract

The challenge of producing sufficient food for a growing population requires increased production efficiency, especially when production is extended into suboptimal production environments. In such areas, resource conservation is increasingly important and innovative linkages between waste streams and food streams can have significant impact on total system efficiency. Bioregenerative Life Support Systems (BLSS) for space have been designed in which plants and mushrooms are grown in tandem, maximizing total food produced, minimizing inedible waste, and maximizing energy and space efficiency. The purpose of this work was to evaluate the application of BLSS for food production on Earth as Bioregenerative Food

Production Systems (BFPS). For this project, a model was created to predict the productivity of a BFPS with production components for plants, mushrooms, insects, and fish. The overall goal was to fulfill the dietary needs of a single person for one day according to the NASA Basile Values and Assumptions Document (BVAD), as well as to feed a specific population size for a specific length of time. Inedible plant material was used to support mushroom production; spent mushroom substrate was used to support insects; and remaining substrate from insect production was used to support fish. To close the energy and resource loop, fish waste (ammonia and suspended solids) was used to supplement input nutrients for plant production. Input for this model relied on data from journal articles, government agencies, and industry websites. All data was accumulated in a cornerstone data page and all component production pages referenced this cornerstone page for calculations. The first page included only plant production components; subsequent components were added onto prior production runs in an iterative process. Each production run was compared to previous and subsequent production runs for water, energy, and growing space efficiencies, and net waste products.

II. Introduction

The world's population is nearly 8 billion and growing.[1][2][3][4] Without substantially revising production, there is not enough arable land to feed the predicted population increase with conventional farming methods. As such, food production is increasingly accomplished using controlled environment agriculture (CEA),[5] which focuses on the environment and environmental inputs of a defined growing space, allowing for the optimizing of plant growth through the manipulating production inputs. Moreover, CEA systems allow for year-round food production, and food production within city limits. Modern greenhouse production is often accomplished via hydroponics, the cultivation of plants using nutrient- and oxygen-rich water instead of soil. In such systems, plants are either supported by a growing substrate, or are suspended on the surface of a nutrient solution (deep-bed culture).[6] Efficiencies of such systems are maximized when production unit are stacked vertically with supplemental lighting in completely enclosed controlled environments.[7]

Most commercial mushroom production also occurs in CEA facilities in which the light, temperature, humidity, and CO2 are closely monitored and tightly controlled.[8] Commercial mushroom production is one of the most valuable specialty crops based upon annual sales and is one of the fastest growing specialty crop commodities. In 2015, commercial market analyses placed world production value at approximately $30 B annually. Coupled with a 10-15% projected annual growth, the industry is positioned to surpass $50-70 B by 2024.[9] As decomposers, mushrooms can be grown on a variety of substrates, including agricultural wastes.[8]

In addition to plant and mushroom production, other areas of CEA include insect agriculture and aquaculture,[10] the cultivation of edible insects and fish, respectively. Edible insects are recognized by the FAO as an important source of protein for human consumption and insect farming, a food industry with exponential growth, is recognized as a priority by the USDA.[11][12] Insects are rich in protein and minerals and require much less resources than do vertebrate livestock.[3][13] Aquaculture, the cultivation of fish and other aquatic lifeforms, has a long tradition of production in open ponds or floating pens, but is increasing conducted in CEA facilities where more precise control can be maintained. The industry is currently experiencing exponential growth with subsequent extension to arid and semi-arid regions around the world.[10]

For enhanced efficiencies, systems can also be linked in CEA facilities. For example, aquaculture can be linked to the cultivation of plants through aquaponics.[14] In aquaponics, the feces produced by the fish are used to sustain the plant crops while also cleaning the water. Aquaponics has several advantages over traditional field agriculture, such as a 90% reduction in water use, reduced use of fertilizers, reduced risk of soil-borne diseases, and the ability to be grown in any location with access to electrical energy.[15] A second focal point has been the development of sustainable aquaculture feeds. Replacing fishmeal in aquafeeds is one of the most critical issues of aquaculture. Insects, fungi, algae, and composted plant materials are some of the key alternative ingredients that have been promoted as fishmeal substitutes and use of aquaculture effluents as nutrient rich organic fertilizer brings together the best parts of the Green and Blue Revolutions.[15]

An extension of CEA is the closed ecological system,[16] in which an ecosystem is sustained in a controlled environment with minimal exchange of resources between the enclosed ecosystem and the external environment. One of the largest examples was Biosphere 2 in Oracle, Arizona. Biosphere 2 was an experiment attempting to sustain 8 people in an enclosed, artificially maintained biosphere for an extended period, as a prototype for space colonization. The concept of sustaining humans using closed ecological systems has been explored several times since Biosphere 2. One such project developed at the University of Arizona is the Mars Lunar Greenhouse (MLGH), which was a greenhouse designed to provide both food and life support for humans on an extraterrestrial colony.[17]

Gellenbeck et al explored the concept of a Bioregenerative Life Support System (BLSS) in 2019.[18] The BLSS initially relied on plant production (like the MLGH), leaving a large amount inedible plant biomass as unusable waste. Gellenbeck's goal was to demonstrate the feasibility of adding an edible mushroom module to the BLSS as a means of converting the inedible biomass to edible mushrooms. He demonstrated that using waste plant material to produce mushrooms would lead to a 33% decrease in growing space and a 31% reduction in electrical consumption.

The purpose of this project is to expand the use of a BLSS for terrestrial food production (now termed a Bioregenerative Food Production System - BFPS) with the inclusion of insect and fish production added to the system in an iterative manner to further utilize system waste streams as resources for alternative food production. These additional components have the potential to further reduce growing space, water use, energy consumption, and waste production while also maximizing food production.

Materials, Methods, and Results.

Database development

This project builds off Gellenbeck et al,[18] and as such, the same initial data inputs, selected plant and fungal species, and nutritional requirements were used to initiate this study. Plant species included sunflower (*Helianthus annuus*), spinach (*Spinacia oleracea*), and quinoa (*Chenopodium quinoa*). The species of fungus selected was the pearl oyster mushroom (*Pleurotus ostreatus*). The human nutritional requirements were obtained from NASA's Baseline Value and Assumptions Document.[19] Based upon these data, a hypothetical crew required the following amounts of nutrients on a per-day, per-crew member basis: 3000 kilocalories, 405 grams of carbohydrates, 123 grams of protein, and 103 grams of fat. The Baseline plant and mushroom data included species utilized, their nutritional profiles, and the inedible portion of the crop remaining after harvest.

This expanded project required species selection for an insect and a fish component to be added to the system. The chosen insect was the larvae of the mealworm beetle (*Tenebrio molitor*), a species of insect commonly used as food for pets and for humans. This species was also chosen for its high protein content; 20% of live weight, 50% of dried weight.[20] The chosen fish was the Nile tilapia (*Oreochromis niloticus*), a species of fish commonly used in aquaculture and aquaponics due to its comparatively rapid growth of 6 months and high stocking density.[21] In an iterative manner, insects were added to consume the spent mushroom substrate (SMS) remaining after mushroom production, and fish were added to consume the insects and remaining unused insect food (SMS) following insect production. Based upon these data, fresh weight of product and its nutrition was calculated on a per person per day basis, but also for a hypothetical crew of 8 (Biosphere 2 population) for 183 days (the longest production cycle among the food components.

Data points and assumptions for this project, as well as applicable formulae, were acquired through review of primary literature, industry websites, and government databases such as that of the Food and Agriculture Organization of the United Nations. Combining this data with additional information such as nutrient content, feed conversion ratios for the animal crops, harvest indices for plants, standard yield values, standard rainfall requirements, and energy requirements, a matrix was created and labeled “Cornerstone Data”. Most of these data are shown in Tables 1a and 1b**. U**nits presented are the basis for the actual calculations; result tables have appropriate unit conversions. Data for sunflower come from or is calculated with data from Gellenbeck et al,[18] González-Pérez,[22] Nielsen,[23] the FAO,[24] and Brouwer et al. [25] Data for spinach come from or is calculated from Gellenbeck et al,[18] Brouwer et al,[25] and Campbell-Nelsen.[26] Data for quinoa come from or is calculated from Gellenbeck et al,[18] Hassoon and Dziki,[27] ProAg,[28] Peterson and Murphy,[29] and Garcia et al.[30] Data for oyster mushrooms come from Gellenbeck et al,[18] and Stamets.[31] Data for mealworms come from Thevenot et al,[32] van Huis,[11] Zhao et al,[33] and Morales-Ramos.[34] Data for tilapia come from Guzman-Luna et al.[35] and Staughton.[36]

Table 1. 1: Nutritional content and requirements for species used in BFPS

Species	Calories (kcal/kg FW)	Carbs (g/kg FW)	Protein (g/kg FW)	Fats (g/kg FW)	WC of organism	WC of harvested material
Sunflower	6250	150	225	575	92%	6%
Spinach	230	36	29	4	92%	92%
Quinoa	3680	642	141	61	92%	11%
Oyster mushroom	330	61	33	4	92%	92%
Mealworm	2060	1	200	150	62%	62%
Tilapia	960	0	200	17	78%	78%

Table 1. 2: Nutritional content and requirements for species used in BFPS

Species	Harvest Index	Time to Harvest (d)	AR (m^2/kg)[1]	WR (L/kg)[2]	ER (MJ/kg)[3]
Sunflower	40%	100	4.5	1019.1	22.25
Spinach	90%	41	0.6	97.0	2.21
Quinoa	30%	105	2.7	549.0	13.52
Oyster mushroom	92%	28	0.1	2.1	0.01
Mealworm	100%	129	0.3	0.6	4.14
Tilapia	37%	183	0.1	112.4	24.45

Production Runs.

Run 1: Plants

In the first production run (Run 1), the selection of crops was based upon those used in Gellenbeck et al 2019. (a correction was made on fresh weights for quinoa and spinach) and the amounts of fresh weight produced would fulfill the BVAD requirements.[18] If the resultant

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[1] Area requirement for plant crops was calculated from standard crop yield data. Area requirements for mushroom crops was calculated from densities from Gellenbeck et al, 2019 and the ability to stack growing shelves three-high. Area requirements for insects were based upon standard densities of 1 kg/m2, and the ability to stack growing containers (h = 0.1 m) three-high. Area requirement for tilapia was calculated based upon standard production density (10 kg/m^3) and a tank depth of 1 meter.

[2] Water requirement for plant crops was calculated based upon standard rainfall/irrigation needs and yield. Water requirements for mushrooms was based upon rehydration requirements of substrates to 70%. Water requirement for insects was to provide sufficient moisture for hydration. Water requirement for tilapia was sufficient to fill tanks for fish production.

[3] Energy requirements for plant and mushroom crops were calculated based upon photosynthetic/illumination requirements using electrical lighting (Gellenbeck et al 2019), time to harvest, and standard crop yields for each crop cycle. Energy requirements for insects was based upon air circulation and temperture requirements and time to harvest. Energy requirement for tilapia was based upon circulation, filtration, and temperature requirements, time to harvest.

nutrients were found to be excessive for or insufficient for the BVAD requirements, the fresh weights were adjusted until the nutrients met or exceeded the BVAD requirements by a small margin. The results for Run 1 are shown in Tables 2.1 and 2.2.

Table 2. 1: Per-person, per-day results for Run 1 (Plants)

Species	FW (g/day)	Calories (kcal/day)[4]	Carbs (g/day)	Protein (g/day)	Fat (g/day)
Sunflower	130	813	19	29	75
Spinach	300	69	11	9	1
Quinoa	605	2226	388	85	37
Total	1035	3108	418	123	113
BVAD Requirements.		3000	405	123	103

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[4] Nutrient calculations use equation 1.

Table 2. 2: Per 8 persons, per 183 days results for Run 1 (Plants)

Species	FW (kg)[5]	AR (m^2) [6]	WR (liters)[7]	ER (MJ)[8]	IPM$_{(WW)}$ (kg)[9]	IPM$_{(DW)}$ (kg)[10]
Sunflower	190	855	193,610	4227	285	23
Spinach	439	263	42,583	970	49	4
Quinoa	886	2392	486,414	11,979	2067	165
Total	1515	3510	722,607	17,376	2401	192

$$Nutrient[Cal, Carb, Prot, or\ Fat](kcal, kg) = FW\left(\frac{kg}{day}\right) * (nutrient\ content)(\frac{kcal,kg}{kg}) \tag{1}$$

$$FW(2b)(kg) = FW(2a)(kg) * 183 * 8 \tag{2}$$

$$AR(m^2) = FW(2b, kg) * area\ per\ kg(\frac{m^2}{kg}) \tag{3}$$

$$WR(plants, L) = AR(m^2) * Water\ needed\ per\ day\ (m) * (\frac{1000\ L}{1\ m^3}) \tag{4}$$

$$ER(GJ) = FW(2b, kg) * energy\ needed\ \left(\frac{kWh}{kg}\right) * (\frac{0.036\ GJ}{1kWh}) \tag{5}$$

$$IPM_{WW}(kg) = FW(2b, kg) * (\frac{1-HI}{HI}) \tag{6}$$

$$IPM_{DW}(kg) = FW(2b, kg) * (1 - WC(\%)) \tag{7}$$

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[5] Fresh weight produced for Table 2b uses equation 2.

[6] Area required uses equation 3 and data from table.

[7] Water needed uses equation 4 and data from table 1, converted to liters.

[8] Energy needed uses equation 5 and data from table 1, converted to Megajoules.

[9] Inedible plant material wet weight uses equation 6 and data from table 1.

[10] Inedible plant material dry weight uses equation 7 and data from table 1

Run 2: Plants and Mushrooms

After Run 1 nutrition calculations, follow-up calculations were conducted to convert the resultant inedible plant material into a substrate for mushroom production. This included addition of water to increase the dry substrate's water content to 70%. No additional water was needed during production except to maintain humidity in the growing area at 80% RH. The fresh weight of mushrooms producible was then calculated using Run 1 inedible plant material (dry weight) and the feed conversion ratio 1.0 (bio-efficiency % = 1/feed conversion ratio) used by Gellenbeck et al.[18] After performing these calculations, the results were included in the calculations for Run 2, rebalancing the plant fresh weights until the total fresh weight provided sufficient nutrition. The results are shown in Tables 3.1 and 3.2.

Table 3. 1: Per-person, per-day results for Run 2 (Plants and Mushrooms)

Species	FW (g/day) [11]	Calories (kcal/day)	Carbs (g/day)	Protein (g/day)	Fat (g/day)
Sunflower	130	813	20	29	75
Spinach	290	67	10	8	1
Quinoa	580	2134	372	82	35
OM	126	42	3	4	1
Total	1126	3055	405	124	112
BVAD Requirements		3000	405	123	103

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[11] OM fresh weight uses equation 8.

Table 3. 2: Per 8 persons, per 183 days results for Run 2 (Plants and Mushrooms)

Species	FW (kg)	AR (m^2)	WR (liters) [12]	ER (MJ)	$IPM_{(WW)}$ (kg)	$IPM_{(DW)}$ (kg)	$SMS_{(DW)}$(kg) [13]
Sunflower	190	855	193,610	4227	286	23	-
Spinach	425	255	41,225	939	47	4	-
Quinoa	849	2292	466,101	11,478	1981	158	-
OM	185	18	370	2	(2314)	(185)	170
total	1649	3420	701,306	16,646	0	0	170

$$FW_{OM}(kg) = sum\ of\ IPM_{DW} * BE(\%) \tag{8}$$

$$DW_{OM}(kg) = FW_{OM}(kg) * (1 - WC_{OM}) \tag{9}$$

$$SMS_{DW}(kg) = SUM\ of\ IPM_{DW}(kg) - DW_{OM}(kg) \tag{10}$$

$$WR_{OM}(L) = SMS(kg) * \left(\frac{70}{30}\right) * \left(\frac{1m^3}{977kg}\right) * \left(\frac{1000L}{1m^3}\right) \tag{11}$$

Run 3: Plants, Mushrooms, and Insects

After Run 2 nutrition calculations, the resulting spent mushroom substrate (SMS) was used to calculate the mass of mealworms producible. This was accomplished by multiplying the dry weight of substrate by the inverse of a feed conversion ratio of 1.8, obtained from Alexander, 2017,[3] as shown in equation 8. No additional water was added to the dry SMS for use as mealworm feed; minimal water supplement was provided by small amounts of fresh vegetable matter. The amount of frass (insect digestive waste) produced was also calculated using an equation involving the fresh weight of the mealworms, the feed conversion ratio for mealworms,

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[12] OM water required uses equation 11.

[13] SMS uses equation 9 and 10

and the estimation of 100% SMS consumption. The results for Run 3 are shown in Tables 4.1, 4.2, and 4.3 (percent carbohydrate for mealworms is considered negligible, but 0.001 was used for calculations).

Table 4. 1: Per-person, per-day results for Run 3 (Plants, Mushrooms, Insects)

Species	FW (g/day)	Calories (kcal/day)	Carbs (g/day)	Protein (g/day)	Fat (g/day)
Sunflower	120	750	18	27	69
Spinach	260	60	9	7	1
Quinoa	577	2123	370	82	35
OM	124	41	8	4	<1
Mealworms	64	131	<1	13	10
total	1145	3105	405	133	115
BVAD Requirements	1145	3000	405	123	103

Table 4. 2: Per 8 persons, per 183 days results for Run 3 (Plants, Mushrooms, Insects) Part 1

Species	FW (kg) [14]	AR (m^2)	WR (liters) [15]	ER (MJ)
Sunflower	176	792	179,344	3916
Spinach	380	228	36,860	840
Quinoa	845	2281	463,905	11,424
OM	182	18	382	2
Mealworm	93	28	56	385
total	1676	3347	680,547	16,567

Table 4. 3: Per 8 persons, per 183 days results for Run 3 (Plants, Mushrooms, Insects) Part 2

Species	IPM$_{(WW)}$ (kg)	IPM$_{(DW)}$ (kg)	SMS$_{(WW)}$ (kg)	SMS$_{(DW)}$ (kg)	AF (kg) [16]
Sunflower	264	22	-	-	-
Spinach	42	4	-	-	-
Quinoa	1971	160	-	-	-
OM	(2277)	(186)	559	168	-
Mealworms	-	-	(559)	(168)	35
total	0	0	0	0	35

$$FW_{MW}(kg) = \frac{SMS\,(kg)}{FCR_{MW}} = SMS(kg) * BE(100\%) \tag{12}$$

$$WR_{MW}(L) = FW_{MW}(kg) * WR_{MW} \tag{13}$$

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[14] Mealworm Fresh weight uses equation 12

[15] Mealworm water required uses equation 13

[16] Insect animal waste, or frass, uses equation 14

$$AF(kg) = FW_{MW}(kg) * \left(1 - \frac{1}{FCR_{MW}(1.8)}\right) \tag{14}$$

Run 4: Plants, Mushrooms, Insects, and Fish (all insects go to fish)

After Run 3 nutrition calculations, the mass of insects produced, and the remaining SMS, were used to calculate the mass of tilapia producible. Tilapia feed requires a protein content of 28-36% (grams of protein per gram DW of feed).[37] Since the dry protein content of the mealworms was calculated to be 52.49%, all the mealworms (100%) were mixed with the uneaten SMS (protein content calculated at 5% for SMS) to achieve a tilapia feed content of 36%.

After calculating the mass of fish food producible, mass was multiplied by the inverse of a feed conversion ratio of 1.6, obtained from Recsetar, 2020,[38] to obtain the weight of tilapia, then multiplied by a harvest index of 37% to obtain the fresh weight of tilapia.[39] The total fish waste produced by the fish was also calculated using equations 19 and 20 from Recsetar, 2020.[38] These calculations were included into Run 4, with a final rebalancing for the plant biomass. The results for Run 4 are shown in Tables 5.1, 5.2, and 5.3.

Table 5. 1: per-person, per-day results for Run 4 (Plants, Mushrooms, Insects, Fish (100% insects to fish))

Species	FW (g/day)	Calories (kcal/day)	Carbs (g/day)	Protein (g/day)	Fat (g/day)	$FW_{(MW)}$ for tilapia (g/day)	$DW_{(MW)}$ for tilapia (g/day)
Sunflower	128	800	19	29	74	-	-
Spinach	260	60	9	9	1	-	-
Quinoa	577	2123	370	81	35	-	-
OM	125	42	8	4	<1	-	-
Mealworms	0	0	0	0	0	47	18
Tilapia	49	46	0	9	1	-	-
total	1139	3071	406	132	111	47	18
BVAD Requirements		3000	405	123	103		

Table 5. 2: Per 8 persons, per 183 days results for Run 4 (Plants, Mushrooms, Insects, Fish (100% insects to fish)) Part 1

Species	FW (kg)	FW_{MW} for tilapia (kg)	AR (m^2)[17]	WR (liters)[18]	ER (MJ)[19]	$IPM_{(WW)}$ (kg)
Sunflower	187	-	841	190,553	4161	281
Spinach	381	-	229	36,957	842	42
Quinoa	845	-	2281	463,905	11,424	1971
OM	183	-	18	384	2	(2294)
Mealworm	0	69	21	41	286	-
Tilapia	71	-	7	7980	1736	-
total	1667		3397	699,800	18,451	0

*Fish area needed uses equations 18 ,19, and 20.

**Fish water needed uses equations 18 and 19

*** Fish energy needed is explained in *Additional calculations*.

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[17] Fish Water required uses equation 17.
[18] Fish Area required uses equation 18.
[19] Fish Energy Required is explained after Tables 6a and 6b.

Table 5. 3: Per 8 persons, per 183 days results for Run 4 (Plants, Mushrooms, Insects, Fish (100% insects to fish) Part 2

Species	$IPM_{(DW)}$ (kg)	$SMS_{(WW)}$ (kg)	$SMS_{(DW)}$ (kg)	$AW_{(WW)}$ (kg) [20]	$AW_{(DW)}$ (kg)	AF (kg) [21]
Sunflower	23	-	-	-	-	-
Spinach	3	-	-	-	-	-
Quinoa	158	-	-	-	-	-
OM	(184)	563	169	-	-	-
Mealworms	-	(417)	(125)	0	0	26
Tilapia	-	(146)	(44)	144	32	28
total	0	0	0	144	32	54

$$TW_{TN}(kg) = \frac{FW_{TN}(kg)}{HI_{TN}(37\%)} \tag{15}$$

$$AW_{TN}(kg) = TW_{TN}(kg) * (1 - HI_{TN}) \tag{16}$$

$$WR_{TN}(L) = TW_{TN}(kg) * WR_{TN}(\frac{L}{kg}) \tag{17}$$

$$AR_{TN}(m^2) = TW_{TN}(kg) * AR_{TN}(m^2/kg) \tag{18}$$

$$TSS(kg) = 0.25 * FF_{DW}(kg) \tag{19}$$

$$TAN(kg) = 0.92 * (\%protein_{FF}) * FF_{DW}(kg) \tag{20}$$

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[20] Tilapia inedible mass uses equations 15 and 16

[21] Tilapia feces is the sum of equations 19 and 20.

Run 5: Plants, Mushrooms, Insects, Fish (50% of insects to fish)

A new production run was calculated to utilize some of the insects for human consumption. This run was identical to run 4, but with the percentage of insects fed to fish being made into a variable, with 50% as the default. With the change in insects fed to fish, the amount of substrate fed to the insects needed to be modified to reach the target protein content of 36%. This was rebalanced to meet the nutritional parameters. The results are shown in Tables 6.1, 6.2, and 6.3.

Table 6. 1: Per-person, per-day results for Run 5 (Plants, Mushrooms, Insects, Fish (50% insects to fish))

Species	FW (g/day)	Calories (kcal/day)	Carbs (g/day)	Protein (g/day)	Fat (g/day)	$FW_{(MW)}$ tilapia (g/day)	$DW_{(MW)}$ (g/day)
Sunflower	128	800	19	29	74	-	-
Spinach	260	60	9	7	1	-	-
Quinoa	575	2116	369	81	35	-	-
OM	125	41	8	4	<1	-	-
Mealworm	23	47	<1	5	3	23	9
Tilapia	26	25	<1	5	<1	-	-
total	1137	3089	405	131	114	23	9
BVAD Requirements		3000	405	123	103		

Table 6. 2: Per 8 persons, per 183 days results for Run 5 (Plants, Mushrooms, Insects, Fish (50% insects to fish)) Part 1

Species	FW (kg)	$FW_{(MW)}$ for tilapia (kg)	AR (m^2)	WR (liters)	ER (MJ)
Sunflower	187	-	841	190,533	4161
Spinach	381		229	36,957	842
Quinoa	842	-	2273	462,258	11,384
OM	183	-	18	384	2
Mealworms	33	33	20	40	274
Tilapia	38	-	4	4271	929
total	1664	33	3385	694,443	17,592

Table 6. 3: Per 8 persons, per 183 days results for Run 5 (Plants, Mushrooms, Insects, Fish (50% insects to fish)) Part 2

Species	$IPM_{(WW)}$ (kg)	$IPM_{(DW)}$ (kg)	$SMS_{(WW)}$ (kg)	$SMS_{(DW)}$ (kg)	$AW_{(WW)}$ (kg)	$AW_{(DW)}$ (kg)	AF (kg)
Sunflower	281	23	-	-	-	-	-
Spinach	42	3	-	-	-	-	-
Quinoa	1965	157	-	-	-	-	-
OM	(2288)	(183)	0.38	168	-	-	-
Mealworm	-	-	(0.27)	(119)	0	0	25
Tilapia	-	-	(0.11)	(49)	65	14	15
total	0	0	0	0	65	14	40

Result comparison tab

After performing the calculations for five production runs, all runs were combined to compare resource use for feeding 8 people for 183 days. Areas of comparison included total growing area, water required, and energy consumption for the total amount of fresh weight produced. A net waste column was also included.

Table 7. 1: Comparison of all production runs

Production Run	FW for human consumption (kg)	AR (m^2)	WR (l)	ER (MJ)	NW (kg wet weight)[22]
Run 1	1,515	3510	722,607	17,376	2,401
Run 2	1,649	3420	701,306	16,646	568
Run 3	1,676	3347	680,547	16,567	35
Run 4	1,667	3397	699,800	18,451	175
Run 5	1,664	3385	694,443	17,592	106

$$NW(kg) = IPM_{WW}(kg) + SMS_{WW}(kg) + AW_{TP,WW}(kg) + AF_{WW}(kg) \tag{21}$$

After performing the calculations for the Results tab, it was decided to create a new data table that showed ratios of resources to fresh weight for each production run, enabling easier comparison of resource use efficiency for the production runs. This is shown in Table 7.2.

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[22] Net waste uses equation 21

Table 7. 2 Comparison of Ratios of Resource-to-Fresh-Weight for all 5 Production Runs

Ratios	AR to FR (m^2/kg)	WR to FR (L/kg)	ER to FR (MJ/kg)	NW to FR (kg/kg)
Run 1	2.33	471.64	11.38	1.58
Run 2	2.08	420.77	10.14	0.34
Run 3	1.99	401.17	9.914	0.02
Run 4	2.04	415.32	11.10	0.10
Run 5	2.03	412.84	10.61	0.06

Discussion

Growing space needed

Growing space requirement decreased from Run 1 to Run 3, increased in Run 4, and decreased again in Run 5. This highlights the space efficiency of oyster mushroom revealed in Gellenbeck et al,[18] and of mealworms revealed by Oonincx and de Boar (2012).[13] This decrease in required space occurs while there is a steady increase in the combined FW, which further highlights the space use efficiency. Space savings can also be achieved by utilizing different crops having different harvest times so that multiple crop cycles can occur within the 183-day period, allowing for the reuse of growing space and overall reduction in needed growing area. Further space efficiencies can be achieved by additional stacking of growing units.

Water consumption comparison

Like area requirements, water consumption decreased from Run 1 to Run 3, increased in Run 4, and decreased again in Run 5. The decrease from Run 1 to Run 3 reveals high water efficiency in the production of mushrooms and insects compared to plant production. While water consumption remained high across all production runs, it is important to note that this is consumed over 183 days. This study did not calculate water needed to maintain humidity levels in the mushroom and insect growing module, or water input to replace evaporation losses of the fish component. Recapture of water loss in the plant production, mushroom, and fish systems would result in further increase in water efficiencies.

Energy consumption comparison

Like area and water requirements, energy usage decreased from Run 1 to Run 3, increased substantially in Run 4, and decreased again in run 5. Considering the increase in fresh weight from Run 1 to Run 2, this supports findings of Gellenbeck and Oonincx & Boer regarding the energy efficiency of mushroom cultivation and mealworm cultivation, respectively.[18][13] The increase in energy consumption with the inclusion of the fish component was due to the necessity of water circulation, filtration, and aeration pumps for fish survival. The energy requirement per kg fish was higher than those for all plant species and much higher than those of mushrooms and insects, the other non-plant protein sources. As such, any replacement of mushrooms- or insect-based protein by fish-based protein would lead to a concomitant increase in energy requirements, as well as area and water requirements. Thus, any efficiencies obtained with modification in aquaculture would have large effects on total efficiencies in this system as it was clearly the costliest of this plant-based bioregenerative food production system.

Net waste production comparison

Net waste decreased substantially from Runs 1 to Run 3. This supports the findings from Gellenbeck et al and Thevenot et al that including mushroom and insect production into most multi-component food production systems will result in substantial resource recovery from waste streams.[18][32] This recovery will also result in an increase in area, water, and energy efficiency and an increase to total FW generated, all while maintaining baseline nutritional outputs requirements for comsumers.

Conclusion

The concept of a Bioregenerative Life Support System used on earth as a Bioregenerative Food Production system is promising and could serve as a prototype for food production in areas challenged by resource limitations, food scarcity, and food insecurity. The inclusion of the mushroom and insect components help decrease resource consumption while leading to a lower net waste produced within the system. In terms of resource consumption, the strategy of growing plants, mushrooms, and insects is the most resource-efficient approach. More work needs to be done for such a system to be fully self-sustaining, either on Earth or for space exploration. Following work could include studies on gas exchange and heat exchange, as well as nutrient input reduction from aquaponic components.

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