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The agricultural biome of Biosphere 2: Structure, composition and function

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Abstract

The agricultural mesocosm of Biosphere 2, known as the Intensive Agricultural Biome (IAB), provided food for the inhabitants of the facility during two periods of material closure between 1991 and 1994 (Mission I, September 26, 1991 to September 26, 1993, eight-person crew; Mission II, March 6, 1994 to September 17, 1994, seven-person crew). The design and operation of the mesocosm and preliminary results for food production of the IAB are described for both periods. The overall rate of crop production for the 0.22 ha area (soil depth of 1 m; soil and atmospheric volumes of approximately 2000 m^3 and 38 000 m^3, respectively) sustained both crews. Overall production rates in Biosphere 2 exceeded those characteristic of fertile agricultural land in the most efficient agrarian communities, despite comparatively lower light levels, lack of insect pollinators and unusually dense insect pests. Crop yields were markedly higher for Mission II than for Mission I due, in part, to experience and improvements based on the first closure. The health of the Biospherians is briefly discussed in the context of a low-calorie (1800–2200 kcal day^{-1} per person for

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Mission I and 2200–2400 kcal day^{-1} for Mission II), nutrient-dense diet characteristic of the Biosphere 2 food paradigm. High productivity and biodiversity were due to many factors including high resolution climate control, hyper-intensive agricultural practices, selection and planting of food crops adapted to humid, tropical and sub-tropical conditions, nutrient recycling, intensive pest management, and the superambient levels of atmospheric CO_2 (concentrations up to 4500 ppmv were reported during the 1991 to 1994 occupations). Radiation use efficiency (RUE) for wheat for both periods and a post-Mission II planting were comparable to RUEs observed in other experimental elevated CO_2 settings such as Controlled Ecological Life Support Systems (CELSS) and Free Air CO_2 Enrichment studies (FACE) even though yields were comparatively lower due to low light levels. Integrated management of pests, soil conditions and agricultural practices were key factors in the sustainability of the IAB resulting in minimization of plant loss due to insect herbivory, nematode infestation and reduction in the quality of IAB soils. The use of soils rather than hydroponic systems for the IAB had significant consequences for CO_2, N_2O and O_2 concentrations in the Biosphere 2 atmosphere and rendered primary regeneration technologies ineffective over the periods of closure. The initial high organic carbon content of the IAB soils prescribed by the designers proved to be the largest single source of CO_2 and the largest sink for O_2. The choice of a soil-based compared to a hydroponic-based agricultural system contributed to the accumulation of N_2O to levels as high as 300 times current ambient levels (approximately 310 ppbv). The IAB of Biosphere 2 has the potential, with system improvement, to be a high-yielding, self-sustaining agricultural mesocosm suited for a variety of research endeavors.

Keywords: Agricultural mesocosm; Elevated CO_2; Closed system research; Biosphere 2; Sustainable agriculture

1. Introduction

Can a synthetic, enclosed, soil-based agricultural field system be constructed and managed to simulate the production ecology of the worlds most intensive and productive agrarian systems? This was the challenge for the designers and operators of the Biosphere 2 agricultural mesocosm. The key outcome for the Intensive Agricultural Biome (IAB) of Mission I and Mission II, in the context of the 'Human Experiment,' was simply that sufficient, but not optimal, quantities of food were produced to sustain the eight Biospherians of Mission I for 2 years and the seven member crew of Mission II for about 6 months (e.g. Glenn et al., 1990; Allen, 1991; Silverstone and Nelson, 1996). The health consequences of the food supply for Mission I have been described by Walford et al. (1992, 1995, 1996). The goal of this report is to examine the structure, composition and function of the IAB in the context of a support facility for human sustainability under conditions of material closure. Where possible, comparisons are made for Mission I and Mission II and for subsequent results.

The key elements of the IAB were: (1) a soil bed of large volume allowing intensive farming of diverse crops; (2) an irrigation system that met the moisture requirements of diverse crops; (3) an animal system that minimized plant waste and provided a source of dietary fat and protein; (4) a wastewater system that supported anaerobic and aerobic subsystems; (5) a lagoon-based wetland farming (i.e. wetland rice paddy) and fish farming system (i.e. *Tilapia*); (6) a water management system linked to the potable water supply and to the general water supply of the whole Biosphere 2 facility; and (7) an air duct system that allowed the atmosphere of Biosphere 2 to be passed through the soil column for purification. The biological elements employed in the function of the IAB included pollinators, beneficial insects and higher vertebrates, cultivated earthworms, biologically active soil and selected cultivars of crop species for consumption. The extent to which these systems functioned successfully as individual components and as an integrated complex was variable and is discussed below. The management practices of the IAB with respect to planting and harvesting, control of insect predation and weeds, application and manipulation of water and nutrients and maintenance of soil characteristics were clearly of critical importance in obtaining optimal crop yields. Effective integrated pest management (IPM) was required; toxic pesticides were not allowed in the facility. The production ecology and crop yields of Mission I and Mission II approaches differed and are discussed.

Biosphere 2 began construction in 1986 and commenced the first closure on September 26, 1991, which lasted 2 years. A transition period of 6 months followed Mission I after which Mission II began on March 6, 1994, and lasted about 6 months. A summary of the history of the facility is provided by several articles in this special issue (Allen and Nelson, 1999; Marino and Odum, 1999). During the transition between Missions, emphasis was placed on addition of food crops, particularly banana and papaya to provide more abundant food resources relative to Mission I as well as on upgrading and expanding existing technical systems. The integrity of the atmosphere appeared to remain largely intact during the transition preceding the start of Mission II based on decreases in sulfur hexaflouride (SF_6) concentrations in the enclosed atmosphere; shortly after Mission II began closure was broken briefly by exchange of air via the lung safety glass and airlocks, resulting in approximately a 15% exchange of inside with outside air. Leak rates for the Biosphere 2 facility during the closure periods were estimated at about 10% per year (Dempster, 1994).

In this paper, comparisons of production ecology, crop productivity and nutritional status of the crews are made using data in the open literature as well as archived data in the historic databases maintained by Biosphere 2. In Section 2, the facility and its physical and mechanical components are described followed by a description of the biological components in Section 3. Section 4 discusses results for crop yields, production rates and nutrition followed by the effectiveness of IPM practices and an evaluation of the regeneration technologies. Section 5 concludes with a discussion of the overall success of IAB food production in the context of a human life support system, insights relevant to CELSS studies and future research potential in the area of sustainable agriculture.

2. Design and description

The IAB of Biosphere 2 was located in the west wing of the facility and included the main agricultural area, orchard, balcony and animal system. The IAB covered about 2000 m^2 ($41 \times 54 \times 24$ m), had an air volume of 38 000 m^3 and a soil volume of about 2000 m^3. The floor plan for the IAB is shown in Fig. 1. The 0.22 ha of growing space which faces the west southwest was available exclusively to supply a diverse, nutritionally adequate diet for eight people year-round. The main agriculture area was divided into 18 plots ranging in size from 40 to 102 m^2. Nine large soil boxes on top of air-return ducts were also used for cultivation and divide the IAB into north and south sections (Fig. 1a). Southern sections of the IAB basement, known as the Terai or PD1, were also used for agricultural purposes. There were three aerobic wastewater tanks (fed by an anaerobic digestor) and eight interconnected growing tanks where wetland rice, *Tilapia* fish, bananas and *Azolla* were cultivated (Fig. 1b). The IAB interacted with the other areas of Biosphere 2 by air circulation which mixed IAB air containing gases such as CO_2, N_2O and CH_4 with system wide air; mixing occurred primarily through the Terai fans located at the interface of the IAB basement and the wilderness basement. Drainage water from the IAB soils was mixed with condensate water for irrigation, or was shipped to the primary storage tank. The IAB structure of Biosphere 2 was engineered specifically to be operated as a sealed environment (Dempster, 1994; Zabel et al., 1999).

2.1. Light

The steel spaceframe of Biosphere 2 contained 6600 panes of double laminated glass primarily covering the wilderness areas and the IAB. Shading by the spaceframe and filtering by glass reduced the effective overall transmittance to 50–55% relative to external clear sky conditions and depending on the exact location in the IAB and the sun angle; similar reductions were reported by Finn (1996). Essentially all ultraviolet light was eliminated. Fig. 2a shows external versus internal light intensity as a function of wavelength for five locations in the IAB. Of particular importance to plants was the photosynthetically active radiation (PAR; usually given in micromoles per square meter per second ($\mu mol\ m^{-2}\ s^{-1}$) in the 400–700 nm wavelength band) incident on the floor of the IAB. Fig. 2b shows the effect of shadows and glass on daily internal light levels based on sensor data. These data clearly illustrate that the location of planting, as much as external light level, dramatically influenced the total amount of light a crop received and thus the resulting yield. Light levels varied considerably in different sections of the IAB with the highest and lowest light levels falling on the south west and north east corners, respectively. Internal light levels were low compared to tropical/sub-tropical areas and the seasonal variation was comparatively high; maximum summer values, typically about 65 mol m^{-2} day^{-1}, were about three times the winter minimum values (Fig. 2c). The total amount of light reaching the floor of the IAB per year was similar to levels found in cloudy, rain shadow tropical lowlands (Knapp et al.,

(a)

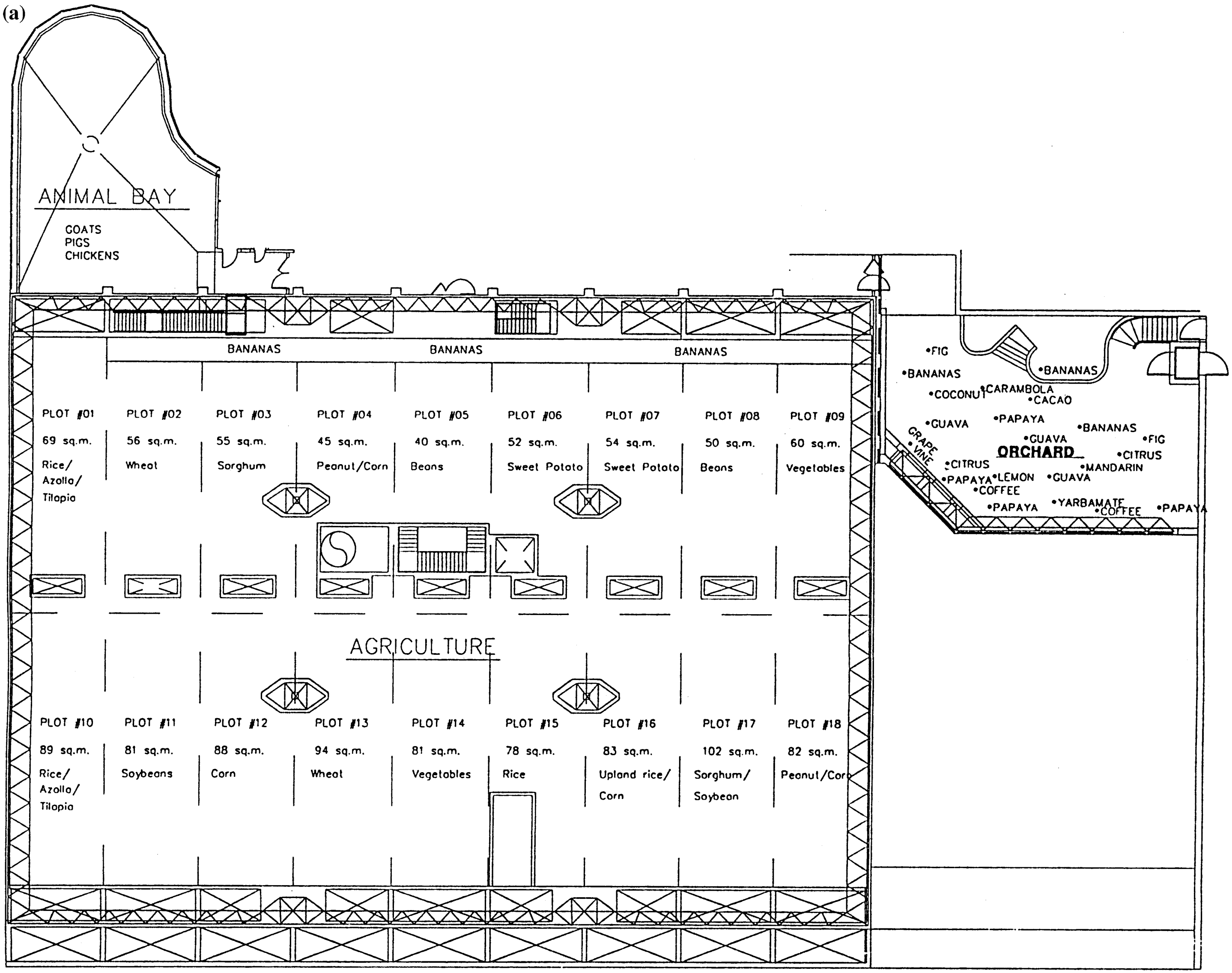

Fig. 1. (*Continued*)

(b)

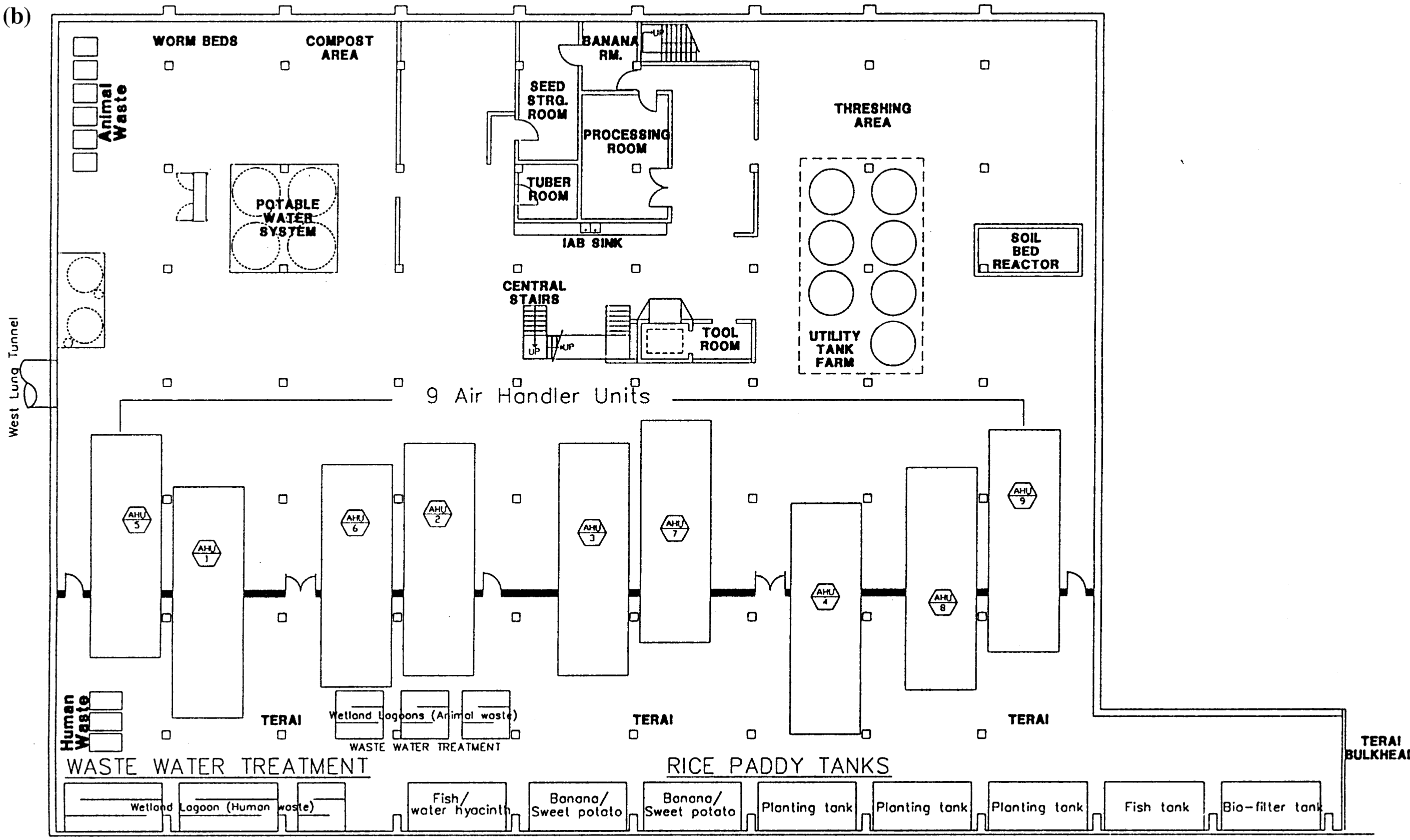

Fig. 1. (a) IAB basement as configured in 1994. Note location of air handlers and separation walls allowing intake of air through Terai (open to upper level planting area) to central air vents and along back wall of the IAB. (b) Floor plan of the planting area of the IAB (including orchard) as it was prepared in 1994. Note animal bay location adjacent to IAB and air handler return vents located in the center of the IAB.

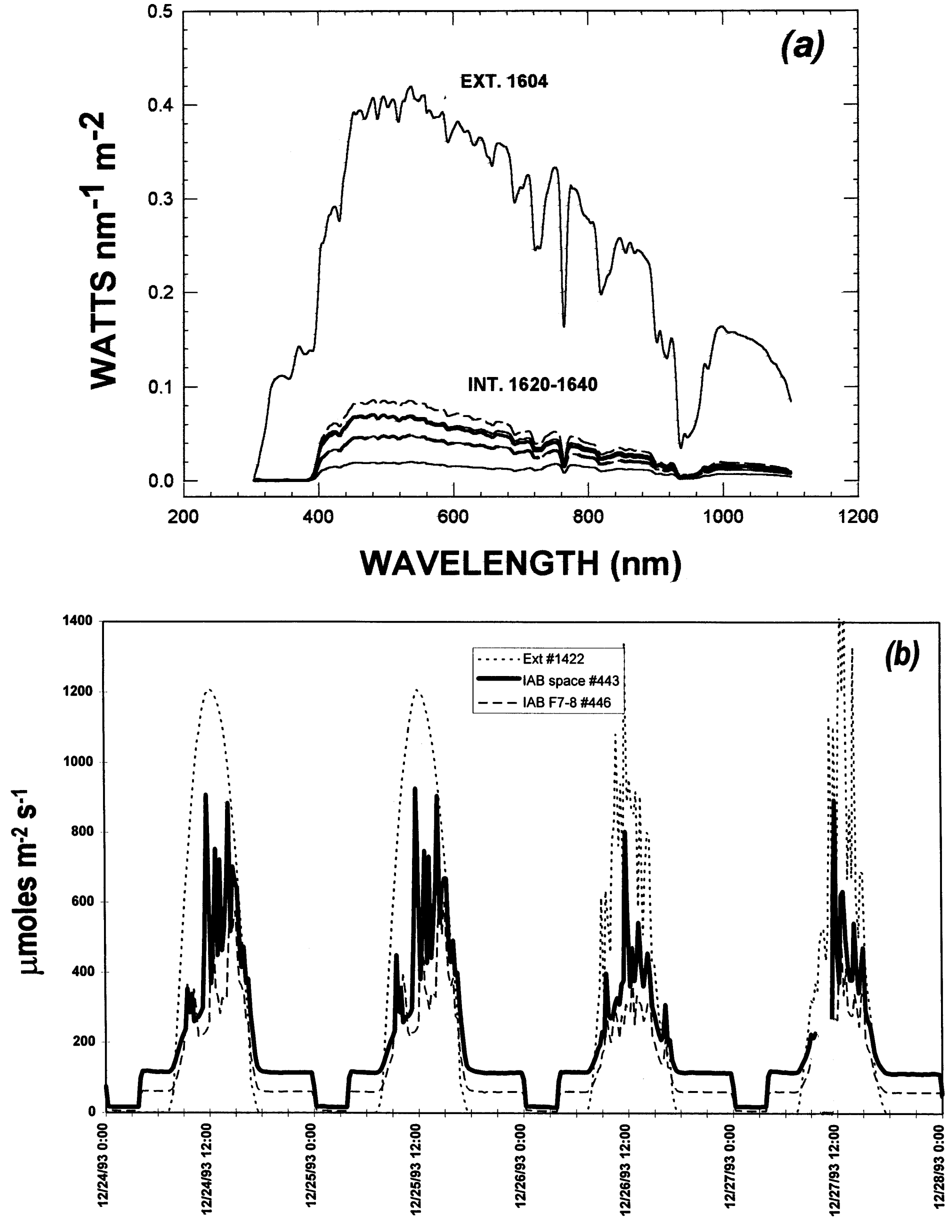

Fig. 2. (a) External and internal irradiance given in W nm^{-1} m^{-2} measured on 5 February 1994, at the times indicated; data for internal locations were selected to reflect maximum and minimum light levels. The measurements were made with a LI-COR LI-1800 Spectroradiometer under clear sky conditions. (b) IAB daily light levels as a function of sensor location. Dotted line represents external light; solid line represents internal light sensor in the space frame; dashed line represents sensor located just above the IAB planting surface showing shadow effects which further attenuated the light field. The contribution of internal lights to plant growth (solid and dashed line for night time intervals), based on available data, was minimal. (c) Seasonal change in external (dotted line) and internal light levels (dashed and solid lines) from November 1993 to May 1995.

1980). While the original design goal was to maximize PAR in the Biosphere 2 facility, multiple spaceframe shadow effects (as seen in Fig. 2b) resulted in a light field that was weaker than anticipated. The reduction in light and the consequences for crop productivity and human health were addressed by the addition of artificial lighting after termination of Mission 1. The artificial lighting system in the IAB agricultural area consisted of 196 fixtures; each contained a 1000-W high pressure sodium lamp. While the original intent of using artificial light was to increase total daily light received, according to available records they were not consistently used due to crop damage in some areas of the IAB, noise and cost. Their contribution to plant growth, based on available records, is estimated to have been minimal.

2.2. IAB soils

The mineral portion of the IAB soil was obtained from a nearby cattle tank (Wilson Pond) consisting of a clay loam soil with about 5% organic matter. The final soil design, by volume, called for 70% Wilson Pond soil, 15% coarse peat, and 15% commercial, weakly composted mulch. As a consequence, the final mix is estimated to have had about 8–10% organic matter (approximately 5% organic carbon) by volume. The organic-rich nature of the soil was specified by some members of the design team of the IAB, according to internal reports, based on the idea that the more organic matter, the better for overall plant vigor and health. The sand fraction of the soil was less than 10% when tested in mid-1995, and consisted almost exclusively of very fine sand. The thickness of the soil column ranged from

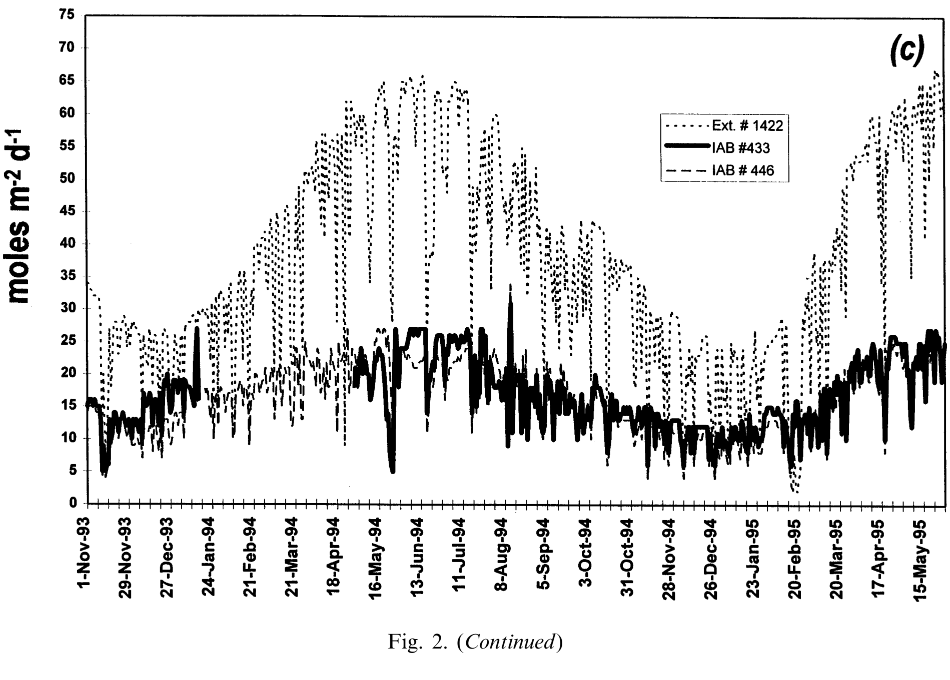

Fig. 2. (*Continued*)

about 80 to 110 cm. The presence of montmorillonite clay in the source soils caused shrinking and swelling; vertisol-type cracks developed which could open up to 5 cm across. In addition, the hard texture of the original IAB soil was difficult to work. The composition and texture of the IAB soils changed throughout the period and are discussed below. The IAB soils eventually became unacceptably saline requiring repeated flushes with clean water as described below. Improvements made in 1994 and 1995 consisted of automatic mixing valves, in-line TDS sensors and the addition of reverse osmosis (RO) water to supplement condensate reserves resulting in high quality water for irrigation of the IAB and the wilderness biomes.

2.3. Soil bed reactor

The IAB (orchard excluded) was built based on the design concept of a soil bed reactor (SBR). The approach is based on decomposition of atmospheric compounds by soil microorganisms (Bohn and Bohn, 1986) or by adsorption as they are introduced by forced air through the soil matrix. The biogeochemistry of soils is complex (e.g. Schlegel, 1974) producing sources and sinks for a variety of naturally occurring and anthropogenically influenced atmospheric gases (e.g. McElroy, 1983; Rheeburgh et al., 1993) and trace contaminants (Bohn, 1972; Khalil and Rasmussen, 1989). The technology has been variously described (e.g. Prokop and Bohn, 1985; Bohn and Bohn, 1988) but not on the scale of the IAB. Preliminary lab and field work to support development of the Biosphere 2 soil bed reactor demonstrated that the effectiveness of such a system would be difficult to control and predict since removal efficiency for a given compound depended on soil moisture, soil composition and physical characteristics, flow rate, plant cover and soil microbial ecology (Frye et al., 1990). The use of the soil bed reactor in an agricultural setting could also result in increased efflux of CO_2, other gases and particulates to the atmosphere from the soil column as well as influence crop productivity; these effects would render the soil bed reactor unsuitable under the prevailing atmospheric composition of high CO_2 and the pressing demand for optimal crop yield. There were 24 sub-soil air vents attached to blowers designed to push 5000 cfm of air into the thin air plenum formed by cinder blocks at the base of the soil. However, the SBR was activated for only a very brief period during Mission I and Mission II for testing and was not used for the purpose of air purification.

2.4. Compost

In the Biosphere 2 approach to life support and sustainability, composting was to serve as the primary vehicle for recycling of solid waste, particularly IAB biomass. However, composting emerged as a questionable practice in Biosphere 2 where the soil was initially rich in nutrients and carbon. Under closed conditions in a system with high atmospheric CO_2 and diminishing oxygen the objective quickly became a reduction in respired CO_2 rather than an increase as occurs during decomposition of compost. Dry storage of biomass was employed instead of allowing compost decomposition. A consequence of not composting was the propagation of weeds, particularly during Mission I, which were added back to the soil in viable form along with shredded biomass.

2.5. Climate control

Temperature and humidity in the agriculture biome were controlled by nine air handler units in the IAB basement that circulated air continuously according to a specified protocol (Fig. 1b). An air handler consisted of coils (water pipes) surrounded by a metal jacket, an intake fan and a controller that modulated air circulation and thermal energy exchange. The air intake end faced south and the exhaust faced the north thereby forcing the air toward the main agricultural area. Hot, chilled, and tower water were pumped through sealed pipelines from the Energy Center in a closed loop (Zabel et al., 1999). The air was sucked in from the basement and upper IAB into the air handler and passed over the heated or cooled coils. The treated air was then delivered to the biome through nine air vents located at the middle of the IAB, one for each air handler (Fig. 1a). Condensation of water occurred in the air handlers fitted for chilled water and was used for irrigation and as a source for potable water. Environmental sensors including light (quantum and pyranometer), temperature and relative humidity sensors were abundant during the period of study, however, the corrosive environment and inadequate maintenance resulted in incomplete records. In addition, up to 15°C difference between temperature and 15% difference between relative humidity sensors depending on location (e.g. spaceframe, balcony, crop level in different locations) within the IAB introduced unavoidable ambiguities in defining operating environmental conditions.

2.6. Water management system

The main reservoirs of water in the IAB were the Utility Tank Farm (UTF), sub-soil tanks, condensate tanks and potable water tanks in the IAB basement, *Tilapia* and *Azolla* tanks and wastewater (human and animal) holding tanks and lagoons in the basement and Terai areas (Fig. 1b). In addition, a large amount of water was also stored within the soil mass and plant tissues. Irrigation water for the IAB was primarily a mixture of condensate water, waste water effluent and sub-soil drainage water. Condensate water was produced and collected from the air handler units in the IAB basement and stored in three 7560 l tanks. The air handler units also supplied potable water to the habitat. After Mission I an 'Allen Bradley' (Allen-Bradley PLC 5/40) computer-based system for switching valves was installed allowing control of each system depending on specified supply and demand. Sub-soil drainage water was collected in 7560 l tanks in the IAB basement that also received treated wastewater from the human and animal lagoons. When all the sub-soil drainage tanks were filled to capacity, the water overflowed to a small plastic trough through an overflow outlet and excess water was pumped to the primary storage in the south lung. Modifications after Mission I allowed for condensate, RO or a mixture of the two, to be used for the IAB irrigation depending upon their production and availability. Primary storage water was used to feed the RO machines to produce a higher quality water for IAB use.

The Terai (Nepalese word for lowland) was intended to be a key wastewater recycling area which consisted of three aerobic human waste treatment lagoons, three animal waste treatment lagoons, five growing tanks, two fish tanks and one biofilter tank (Fig. 1b). Three aerobic waste water treatment tanks in the Terai contained various plants that were grown for the purpose of water purification after anaerobic digestion of solids (fecal matter) and production of animal fodder based on the general approach of Wolverton (1980). In this approach, human and animal feces and urine are made directly available for aquatic plant growth (e.g. water hyacinth) and other lagoon based plant communities (urine and feces are often separated before treatment in these systems). This was to play an important role in the recycling system since we estimate that at least 30 kg of nitrogen from human waste per year were produced by eight persons; the inclusion of animal waste greatly increased nitrogen input. Few data are available to assess the efficacy of the wastewater treatment system (Nelson et al., 1994, 1999); however, as discussed in Section 4, the level of nitrate in the primary storage water and in the IAB irrigation water rose dramatically during the periods of closure indicating inadequate control of nitrogen recycling. The eight connected tanks were used for production of rice, banana, *Tilapia* and *Azolla*, which were fed to the fish and chickens. As of 1996, the main aquatic plant species were *Canna edulis*, *Ipmoea aquatica*, *Eichhornia crassipes*, and *Typha* spp. Biomass from these lagoons was harvested for animal fodder or added to the soil after shredding. The potable water system which included an ozone/hydrogen peroxide treatment step, also located in the IAB basement, produced water of variable quality as discussed below.

3. Biotic components of the IAB

3.1. Cultivar choices for Mission 1 and Mission 2

As a self-sustaining life support system the goal of the IAB design and management was to support up to 10 resident crew members for potentially indefinite periods. A key factor in the success of this goal was the selection of crop species and cultivars for high yield and adaptability. Cultivars were acquired and tested, first in the on-site greenhouse complex and/or in the Environmental Research Laboratory, University of Arizona, and then inside the IAB. By the time of the first closure the IAB had already been planted and several crops harvested to allow the incoming resident crew immediate sustenance and a functioning, established agricultural system (Silverstone and Nelson, 1996). Many factors were considered in cultivar selection including dietary requirements for protein, fat and carbohydrate as well as an adequate supply of minerals and nutrients; USDA minimum daily requirements were used as guidelines. High yielding cultivars of staple plants were chosen for the limited planting area. Resistance to disease and pest infestation was paramount since the high humidity and temperature were ideal conditions for serious and devastating insect predation. In addition, early maturity was an important consideration since it was desirable to achieve three to four crops per planting area per year. A wide variety of crops were desirable in order to add variety to the kitchen table. The cultivar list for Missions I and II are given in Tables 1 and 2, respectively. Foods grown during the missions were comprised of cereal grains, starch crops, legumes, fruits and vegetables. A limited amount of meat, eggs, milk and fish were also produced from the animal system and by aquaculture. The diet was largely vegetarian, calorically restricted and of low-fat content for both missions (Walford et al., 1992).

3.2. Integrated pest management plan

Pest management was an integral part of crop production. Since Biosphere 2 was a small-scale sealed environment, use of toxic chemical compounds was strictly prohibited. The challenge for Biosphere 2 agriculture was to maintain plant health in the face of little or no wind (needed for pollination), high temperature and humidity (an ideal environment for insect pests) and no synthetic pesticides. Several practices were adopted based on the use of pest resistant cultivars, crop rotation, the introduction of beneficial insects, biological control, environmental manipulation, human intervention and the use of emulsions of natural products and water (called safe sprays). Crop cultivation on a rotational basis with diverse crops was envisioned as one of the most important features of a long-term sustainable system.

4. Results and discussion

4.1. Yields for Mission I and II

From September 1991 to September 1994 the major foods for human consumption were banana, lab-lab bean, beet, cabbage, carrot, eggplant, papaya, rice, wheat, sorghum, squash and sweet potato. The crop yield data show that the yields for Mission II were consistently higher than those for Mission I. Rice, wheat, sorghum, and beet root yields increased by 208, 94, 120 and 343%, respectively during Mission II (Fig. 3a). The corn crop failed during Mission I whereas the Mission II yield proved to be one of the most impressive and successful crops for Biosphere 2. One exception, as seen in Fig. 3a, was the yield for soybean. Soybean yield for Mission II was 18% lower than the yield for Mission I due to increased spacing of the soybean plants to allow inter-cropping with sorghum plants. However, sorghum grain yields for Mission II exceeded those of Mission I (by about 120%) even under inter-cropping conditions.

During Mission I there were increasing difficulties with production as the Mission progressed. Increased pests and disease organisms caused decreases in yields for many of the crops. Wheat and sorghum yields were significantly diminished; wheat by *Pythium* spp., a fungus, and sorghum by a fungus that has not yet been identified. Other problems included broadmite on white potato, cowpea, and

Table 1
Crops grown in the IAB of during the First (1991)–1993) and Second (1994) Closed Missions of Biosphere 2

Scientific name	Common name	Mission One	Mission Two
A.			
Oryza sativa	Rice	Y	Y
	M103		Y
	M201		Y
	Upland rice		Y
Triticum aestivum	Wheat	Y	Y
	Roja		
Sorghum bicolor	Sorghum	Y	Y
	DK 28 E		Y
	X335		Y
Zea mays	Corn	Y	Y
	Dent corn		Y
	471		Y
	512		Y
	564		Y
	Sweet corn		Y
	Pop corn		Y
B. Starch crops			
Ipomoea batatas	Sweet potato	Y	Y
Solanum tuberosum	White potato	Y	Y
Colocasia esculenta	Taro	Y	Y
Dioscorea alata	Yam	Y	Y (No edible yield)
C. Legumes			
Glycine max	Soybeans	Y	Y
	Rillito		Y
	Black jet		Y
	4301		Y
	Envy		Y
	Butter bean	Pole bean**	Y
Pisum sativem	Pea	Y	N
	Beans	Bush bean**	Y
Lab lab purpureus	Lab Lab	Y	Y
Phaseolus sp.	Lima	N	Y
Phaseolus sp.	Pinto	Y	Y
Phaseolus sp.	Navy	N	Y
Phaseolus vulgaris	Kidney	N	Y
Phaseolus sp.	Slender wax	Long tom bean**	Y
Vigna sinensis	Cowpea	Y	Y
Vigna sesquipedalis	Yardlong	Y	Y
	Rice bean	N	Y
D. Oil seed crop			
Arachis hypogaea	Peanut	Y	Y
E. Fruits			
Musa acuminata	Banana	Y	Y
Carica papaya	Papaya	Y	Y
Psidium guajava	Guava	Y	Y
Ficus carica	Fig	Y	Y

Table 1 (Continued)

Scientific name	Common name	Mission One	Mission Two
Citrus aurantiifolia	Lime	Y	Y
Citrus limon	Lemon	Y	Y
Citrus reticulata	Orange	Y	Y
Ananas comosus	Pineapple	Y	Y
Fortunella japonica	Kumquat	Y	Y
Malus pumila	Apple	Y	Y (hardly any yield)
Fragaria sp.	Strawberry	Y	Y (No harvest)
Musa sp.	Plantain	Y (Probably from TRF)	N
F. Vegetables			
Lycopersicum esculentum	Tomato	Y	Y
Beta vulgaris	Beets	Y	Y
Dacus carota	Carrots	Y	Y
Brassica sp.	Mustard leaf	N	Y
Beta vulgaris var.cicla	Swisschard	Y	Y
Brassica oleraceae acephala	Kale	Y	N
Brassica oleraceae caulorapa	Kohlrabi	Y	N
	Oriental greens	Y	Y
Lactuca sativa	Lettuce	Y	Y
Raphanus sativus	Radish	Y	Y
Brassica rapa	Turnip	Y	Y
Allium cepa	Onion	Y	Y
Allium sativum	Garlic	N	Y
Brassica oleraceaebotrytis	Cabbage	Y	Y
Abelmoschus esculentus	Okra	N	Y
Solanum melongena	Egg plant	Y	Y
Luffa cylindrica	Sponge gourd	N	Y
Cucurbita pepo	Summer squash	Y	Y
Cucurbita maxima	Winter squash	Y	N
Cucurbita moschata	Pumpkin	N	Y
Cucumis sativus	Cucumber	Y	Y
	Rambo		
	Pickling		
Brassica sp.	Pak choi	Y	Y
Lagenaria siceraria	Bottle gourd	N	Y
Cucurbita pepo	Zucchini	N	Y
Capsicum annum	Bell pepper	Y	Y
Capsicum sp.	P. pepper	Y	Y
G. Herbs, spices, beverages			
Cymbopogon citratus	Lemon grass	Y	Y
Origanum sp.	Oregano	??	Y
Mentha spp.	Mint	Y	Y
Ocimum basilicum	Basil	??	Y
Coriandrum sativum	Coriander	??	Y
Petroselinum crispum	Parsley	??	Y
Capsicum frutescens	Chili pepper	Y	Y
Rosmarinus officinalis	Rosemary	??	Y
Allium schoenoprasum	Chives	Y	Y
Foeniculum vulgare	Fennel	??	Y
Ginger officinale	Ginger	??	Y
Curcuma longa	Turmeric	Y	N
Coffea arabica	Coffee	Y	Y
Ilex paraguayensis	Yarbamate	Y	Y
H. Miscellaneous			
Saccharum officinarum	Sugar cane	Y	Y (No harvest)
Manihot esculenta	Topioca	N	Y (No harvest)

Table 2
Plants of the IAB orchard, terai and balcony

Scientific name	Common name	Mission One	Mission Two
A. Orchard			
Eriobotrya japonica	Loquat	Y	N
Ficus carica	Fig	Y	Y
Carica papaya	Papaya	Y	Y
Citrus spp.	Citrus	Y	Y
Psidium guajava	Guava	Y	Y
Cocos nucifera	Coconut	Y	N
Theobroma cacao	Cacao	Y	N
Averrhoa carambola	Star fruit (carambola)	Y	N
Musa spp.	Banana	Y	Y
Vitis sp.	Grape	Y	N
Ananas comosus	Pineapple	Y	N
Beverages			
Coffea arabica	Coffee	Y	Y
Ilex paraguayensis	Yarbamate	Y	Y
B. Main balcony			
Malus pumila	Apple	Y	N
Musa acuminata	Banana	Y	Y
Citrus spp.	Citrus	Y	Y
Ficus carica	Fig	Y	Y
Vitis sp.	Grape	Y	N
Fortunella japonica	Kumquat	Y	N
Carica papaya	Papaya	Y	Y
C. Terai			
Malus pumila	Apple	Y	N
Malpighia glabra	Acerola	Y	N
Musa acuminata	Banana	Y	Y
Theobroma cacao	Cacao	Y	N
Annona muricata	Prickly custard apple	Y	Y
Ficus carica	Fig	Y	Y
Psidium guajava	Guava	Y	Y
Citrus paradisi	Grape fruit	Y	Y
Fortunella japonica	Kumquat	Y	N
Citrus sinensis	Orange	Y	Y
Carica papaya	Papaya	Y	Y
Laurus nobilis	Bay tree	Y	N
Beverages			
Coffea arabica	Coffee	Y	Y
Camellia sinensis	Tea	Y	Y
Ilex paraguayensis	Yarbamate	Y	Y
D. Main IAB			
Carica papaya	Papaya	Y	Y

some beans; nematodes on tomato; spider mites on soybeans and other beans; and aphids on most crops. Because of these problems, soybean, kidney bean, pinto bean, and cowpea were dropped from the planting plans and replaced by the lab-lab bean which were more resistant to these insects. Also, *Leucaena* was dropped in favor of sweet potato, the latter providing both fodder for the animals and a root crop for human consumption. Thus, over time, agricultural diversity diminished with the cultivation of fewer and fewer crops to the extent that by the last quarter of Mission I, approximately 65% of the agricultural area was planted in only three crops: beet, sweet potato, and lab-lab bean. IAB management for Mission II, based on the experiences of Mission I, successfully focused on crops and practices that greatly improved crop yields and diversity.

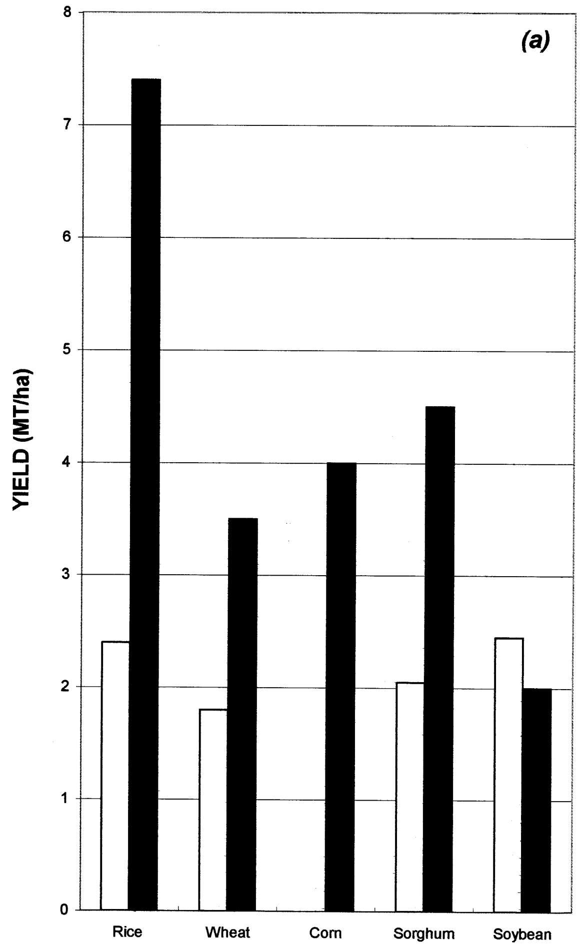

Fig. 3. (a) Selected crop yields for Mission I (unfilled bar) and Mission II (filled bar) given in metric tons per ha (MT/ha). (b) Comparison of rice yields for Asia (Cassman and Harwood, 1995) and for Mission I and II.

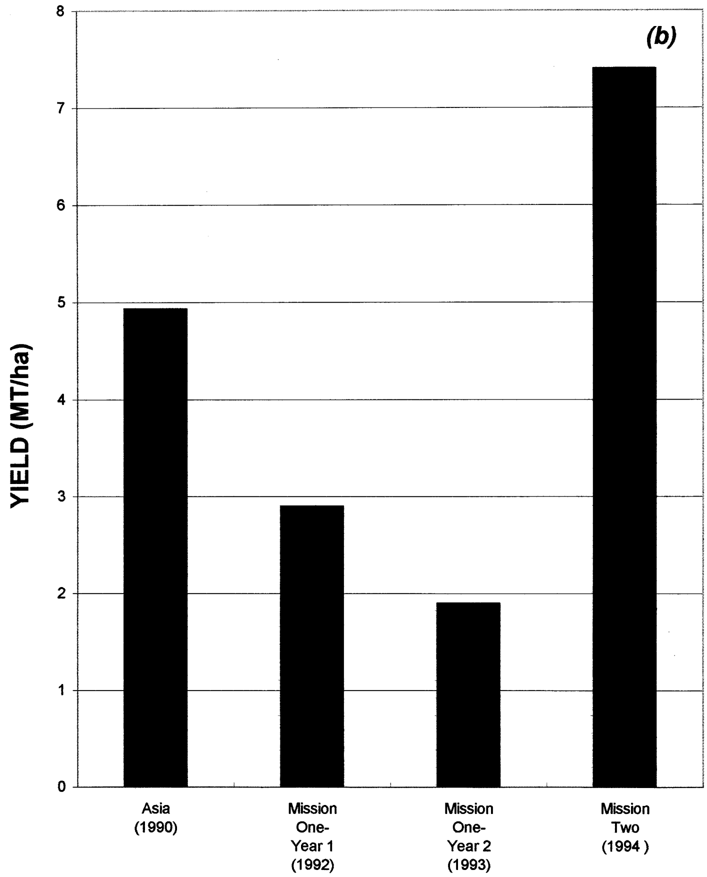

Fig. 3. (*Continued*)

Based on available data, yields for rice for Mission II were higher than average rice yields for the most efficient agrarian communities of Indonesia, Southern China, and Bangladesh (Cassman and Harwood, 1995) (Fig. 3b). In general, total agricultural production in these areas supports a maximum of three persons per acre while the IAB, for both periods of closure, provided food for seven to eight persons on just over half an acre (e.g. Harwood, 1993). Yields for the IAB were all the more dramatic given that the IAB was operated for a short period of time and with comparatively lower light; it seems reasonable to assume that higher yields may yet be possible with additional refinement of production ecology and system improvement. Foliar gas exchange for several agricultural species and observed yields for wheat grown under conditions similar to those of Missions I and II support this view. Results for an experiment with spring wheat carried out in the 1995–96 winter season are described below.

Two C_3 agricultural species (wheat and beans) and one C_4 species were intensively studied during growth under changing CO_2 concentrations (350 to about 2000 ppmv) from February 25 to June 10, 1995 to assess plant physiological function under IAB growing conditions similar to those experienced during the two periods of closure. Photosynthetic CO_2 assimilation, stomatal conductance, ratio of internal to atmospheric CO_2 concentration (Ci/Ca), water use efficiency (WUE) and dark respiration were measured on mature leaves of the selected species with a LI-6200 portable photosynthesis system. The C_4 perennial grass (*Pennisetum purpuretum*) showed no significant change in assimilation rate when CO_2 increased from 350 to 2000 μmol mol^{-1}. However, wheat (*Triticum aestivum*) increased assimilation rate when CO_2 changed from 350 to 1000 μmol mol^{-1}, up to approximately 40 μmol m^{-2} s^{-1}. Pinto bean (*Phasolus vulgaris)* showed a linear increase in assimilation rate over the entire range from 350 to 2000 μmol mol^{-1} suggesting that photosynthetic rate did not reach saturation. The photosynthetic rates achieved for *T. aestivum* were higher than those reported for wheat in at least one study (e.g. Wheeler et al., 1996) and similar to leaf photosynthetic rates measured in NASA-CELSS growth chambers under comparable CO_2 concentrations (Bugbee and Monje, 1992). The saturation concentration of CO_2 was similar at approximately 1200–1500 μmol mol^{-1} in the IAB and in typical CELSS studies (Wheeler et al., 1996). The average enhancement in CO_2 assimilation rate for a doubling of CO_2 from 350 to 700 μmol mol^{-1} observed for C_4 and C_3 plants grown in the IAB was comparable to that found in previous studies. For example, Greer et al. (1995) found a 5% increase in assimilation for selected C_4 species and a 37% increase in assimilation for selected C_3 species in a pasture. The comparability of physiological results for both field and IAB grown plants suggests that the unusual conditions of the IAB (i.e. varying CO_2, relatively low light, high nutrients and large diurnal change in CO_2 concentrations) did not significantly affect plant growth at the leaf level; however, effects on plant development and reproduction are not known (e.g. Bugbee et al., 1994; Grodzinski et al., 1996). The wide variety of plants with species-specific responses to CO_2 levels support the conclusion that high biodiversity was advantageous to increased crop growth given that the levels of CO_2 and other environmental conditions varied considerably. C_3 plants, as expected, were the benefactors of increased CO_2 while C_4 plants showed modest increases in CO_2 assimilation. Maize and sorghum were the only C_4 plants grown in the IAB with high carbohydrate value. High CO_2 levels, as a consequence of the high organic carbon concentration of the soil, was advantageous for a wide variety of C_3 crop species and explains, in part, the high yields observed for the IAB relative to crops grown in the field at ambient levels of CO_2 (approximately 360 μmol mol^{-1}). Bugbee et al. (1994) reported that increasing CO_2 from 340 to 1200 ppmv increased seed yield by 30–40%, consistent with observations for the IAB plants.

The low light intensity field of the IAB compared to ideal field conditions and high-productivity CELSS growth chambers was an important limitation to crop growth and yield. As discussed earlier, the amount of light that reached the plants in Biosphere 2 was attenuated by as much as 55% relative to external light. Further reductions in light occurred based on the exact location and timing of planting in the IAB. These factors make the determination of light conversion efficiencies and other indices of crop productivity related to the light field difficult. However, data for solar radiation from nearby agricultural stations were available to assess changes in regional patterns of sunlight; these data provided an estimate only of the average light striking the glass of the Biosphere 2 facility and cannot account for local weather patterns. Solar radiation data from Tucson (lat., 32°16′49È; long., 110°56′45È; alt., 2340′), Marana (lat., 32°27′40È; long., 110°14′00È; alt., 1973′) and Eloy (lat., 32°46′26È; long., 111°33′25È; alt., 1513′) are assumed to represent average regional cloud conditions for the area, including the Biosphere 2, Oracle, location (lat., 32°34′43È; long., 110°51′21È; alt., 3900′), and are illustrated in Fig. 4. These data clearly show reductions in light during the years 1992 and 1993, however, relative to 1994 the reductions in total annual solar radiation were about 6 and 5%, respectively. The total light received in 1993 was about 4% higher than that of 1992. Monthly reductions in light from one year to the next were variable over the years 1990 to 1995, thus, while light levels during 1992 and 1993 were lower than the preceding and following years, reductions in crop yields were also a consequence of other factors. Based on the data available in Silverstone and Nelson (1996) (Table 2), year one crop yields were about 20% higher than in year two even though light levels were lower in year one than in year two; 12 of the 17 crops common to both years had higher or nearly the same yields for year one. These data support the view that light differences alone do not explain the variation in yields (cf. Silverstone and Nelson, 1996). Silverstone and Nelson (1996) have not cited data to support their claim that El Nino conditions caused above-normal cloudiness during 1991 and 1992. We note that El Nino conditions occurred in

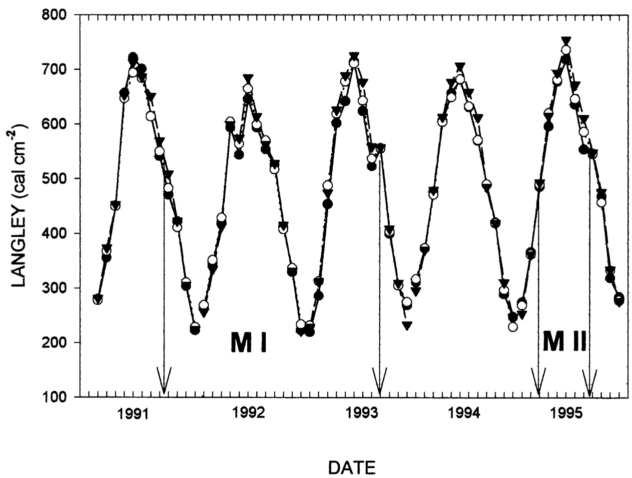

Fig. 4. Light levels shown in Langleys (cal cm^{-2}) obtained from the Arizona Meteorological Network for the stations of Tucson (filled circle), Eloy (filled triangle) and Marana (open circle). Time periods for Mission I and II are indicated.

1991–1992 and 1994–1995 (Kumar and Hoerling, 1997). In any case, simulations with a wheat model parameterized for IAB conditions indicated that differences of 5% in solar radiation among growing seasons were unlikely to generate large differences in crop yields.

Given the scarcity of data on light quality inside the IAB, a valid approach to analyzing crop yields in Biosphere 2 was the integration of observed data and model simulations. In a companion paper, Tubiello et al. (1999) present simulations of wheat data relative to a 1995 growth experiment in the IAB with *Yecora Rojo*, a spring wheat cultivar used in FACE studies by Kimball et al. (1995). Data analyses with the model suggest that, although yield levels in Biosphere 2 were limited by low total light integrals, photosynthetic efficiencies inside the IAB, calculated as total dry matter production per unit absorbed radiation (i.e. g mol^{-1} photon), or radiation use efficiency (RUE), were in fact intermediate between optimal field conditions and NASA-CELSS growth chambers (Tubiello et al., 1999). The same model simulations suggest that the important factors increasing photosynthetic efficiency inside the IAB were higher than ambient CO_2 concentrations (in agreement with our previous discussion), and the ratio of diffuse-to-direct light falling above the crop canopy. This suggestion has never been formulated for the IAB, although it is known that the photosynthetic efficiency of crops grown in greenhouses characterized by high diffuse light fractions is often higher than in the field (Sinclair et al., 1992). The wheat yield was more than 50% lower than optimal field growth data relative to the same cultivar, *Yecora Rojo*, collected at the Free-Air CO_2 Enrichment (FACE) experiment station at Maricopa, AZ, during the same period due to low light characteristic of the IAB environment. Actual crop yields and rates of production for the IAB and typical CELSS and FACE experiments are given in Table 3. Thus, the growing environment of the IAB, as constructed and managed, successfully supported plant growth with RUEs similar to those of hydroponically-based systems typical of CELSS and soil-based field approaches as used in FACE experiments but yields were likely to have been higher with an increased light field. A different space frame design, higher transmittance glass and large reflective panels could improve the light field significantly.

Intensive food production techniques employed in the IAB during the Missions were multiple cropping rather than mono-cropping, cereal–legume inter-cropping (see Fig. 5 for views of the IAB) mixed cropping, polyculture and immediate cultivation of a new crop after the previous crop was harvested. Higher crop yields for Mission II resulted from improved technical knowledge based on Mission I experiences, practical field experience, correct cultivation methods and effective artificial pollination. Simple but important details of planting at the best time, land preparation, proper plant spacing, suitable irrigation methods and weed management figured largely in the successful crop production of Mission II. For example, rice and sorghum grain yields were greatly improved by shaking the inflorescence with a stick facilitating the pollination process even though they are self pollinated crops. Cultivation of corn was impossible without artificial pollination because the IAB lacked sufficient wind and pollinators (pollinators introduced for Mission I and later for Mission II did not survive); plant spacing and ground irrigation

Fig. 5. Planting views of the IAB. Upper panel: Cereal/legume based intercropping. Lower panel: View of IAB from balcony in July 1994.

Table 3
Comparisons of plant productivity for selected IAB and CELSS crops[a]

	Wheat	Rice	Corn	Sorghum
Mission I				
Year One[b]				
Yield (kg m^{-2})	0.22	0.29	0	0.24
Growing period (days)	127	121	0	134
Production rate (kg m^{-2} day^{-1})	0.0017	0.0023	0	0.0017
Year Two[c]				
Yield (kg m^{-2})	0.14	0.2	0	0.17
Growing period (days)	108	121	0	134
Production rate (kg m^{-2} day^{-1})	0.0012	0.0016	0	0.0016
Mission II[d]				
Yield (kg m^{-2})	0.35	0.74	0.4	0.45
Growing period (days)	115	122	102	116
Production rate (kg m^{-2} day^{-1})	0.003	0.006	0.0039	0.0038
Post-Mission II[e]				
Yield (kg m^{-2})	0.479	NA	NA	NA
Growing period (days)	133			
Production rate (kg m^{-2} day^{-1})	0.0036			
CELSS				
Yield (kg m^{-2})		0.76 (S.D. 0.18)	NA	NA
Growing period (days)		86		
Production rate (kg m^{-2} day^{-1})		0.0097 (S.D. 2.7)		

[a] Wheeler et al. (1996), data represent four growth experiments under continuous or 20-h photoperiods with PPF of 509–930 μmol m^{-2} s^{-1} and CO_2 concentrations of 1000–1200 ppmv.

[b] CO_2 concentrations ranged from a maximum of approximately 3200 ppmv to a minimum of 1200 ppmv.

[c] CO_2 concentrations ranged from a maximum of approximately 4000 ppmv to a minimum of 2000 ppmv.

[d] CO_2 concentrations ranged from a maximum of approximately 3500 ppmv to a minimum of 850 ppmv.

[e] See text for experimental details.

methods were crucial for successful corn cultivation. Corn failure during Mission I occurred, apparently, due to plant crowding, washing off of the pollen grains by the overhead watering system and experience with the artificial pollination process. Clearly, higher yields for Mission II were also due to effective integrated pest management. Crop damage by insect pests was minimized during Mission II compared to Mission I as described below. Based on the available records the crew of Mission II expended about 50% of the crew time used for agriculture by the Mission I crew, in part, by employing improvements in the operation and function of the IAB based on previous experience.

4.2. Nutrition for Mission I and II

The IAB provided sufficient nutrients to sustain the inhabitants of Mission I and II demonstrating that the IAB design and management was high yielding in the context of human sustainability. Most all of the inhabitants were taking vitamin and mineral supplements on at least an intermittent basis. Thus, it is not known if the plants grown in Biosphere 2 alone would have provided adequate amounts of micronutrients (vitamins and minerals, phytochemicals), since the plants were grown under altered light (i.e. no UV), soil and gaseous environmental conditions. An analysis of the nutrient content of foods grown in Biosphere 2 was, to our knowledge, never undertaken. While large differences in proximate composition between Biosphere 2 grown and field grown plants are not expected, subtle differences could be important over long periods of time. Plants grown under various CELSS conditions (hydroponic growth) were found to have minor differences in proximate composition (Wheeler et al., 1994).

The nutritional and health status and summary of the medical problems encountered for Mission I have been described by Walford et al. (1992, 1995, 1996). According to these studies, the inhabitants of Mission I adapted to a low-calorie (1800–2200 kcal day^{-1} per person) diet by weight reductions of 18 and 20% for males and females, respectively (Walford et al., 1992). Most of the weight loss occurred during the first 6 months of enclosure, when caloric intake averaged 1780 cal (cf. Silverstone and Nelson, 1996). Nutrients other than calories were reported as nutritionally adequate, although as stated above, the inhabitants were also ingesting vitamin and micronutrient supplements. Weight loss occurred in every member of the crew by 8 months after closure and did not substantially increase thereafter even though caloric intake increased to about 2000–2200 kcal day^{-1} per person from the initial levels of about 1780 kcal day^{-1} per person. Concurrent with weight loss and body mass index (weight/(height)^2), lower serum cholesterols, systolic and diastolic blood pressures, leukocyte counts and blood sugars were also observed, similar to observations reported in animal experiments and human starvation experiments (Walford et al., 1992; Hoffer, 1994).

In the second group of inhabitants, rudimentary food intake data were obtained while inside the facility allowing for estimates of group averages of nutrient consumption. Specifically, the harvested crops were weighed and the amount remaining in bulk was also weighed. However, plate waste was not accounted for and data on food intake of individual inhabitants were not collected. Thus, only crude estimates of macronutrient and micronutrient intakes from food could be made. Caloric intakes were adequate to maintain body weight for Mission II inhabitants after an initial, small weight loss during the first several weeks of enclosure. They were also taking variable amounts of vitamin and mineral supplements; no strict protocol was followed. Fig. 6a shows monthly average calorie, protein and fat consumption for the crews of both Mission I and Mission II (data were obtained from the Biosphere 2 data archive); Fig. 6b shows data for body weight of both groups. Less weight loss occurred in Mission II inhabitants (about 5.2%) compared to Mission I inhabitants (about 18–20%) primarily due to better crop variety and production.

One unique nutritional metabolic problem arose in Biosphere 2 which was caused by high levels of atmospheric nitrous oxide (N_2O). N_2O oxidizes methyl cobalamin and deoxyadenosyl cobalamin (active vitamin B12 forms) through oxidation of the cobalt atom and irreversibly inactivates methionine synthase (Koblin et al., 1981). In vitamin B12 deficiency blood homocystine and methylmalonate levels become elevated in blood and can be measured (Selhub et al., 1993). Blood homocystine and methylmalonate levels were measured in the Mission II inhabitants near the time of departure from the facility, after about 5 months (blood was drawn by R.R. during a 1-week stay with the Biospherians along with BDVM). Elevated methylmalonate was found in one individual indicating tissue vitamin B12 deficiency. It should be noted that almost all of the Mission II group were taking at least occasional vitamin supplements which contained vitamin B12. The elevated methylmalonate was found in an individual who was only rarely taking a vitamin supplement containing vitamin B12.

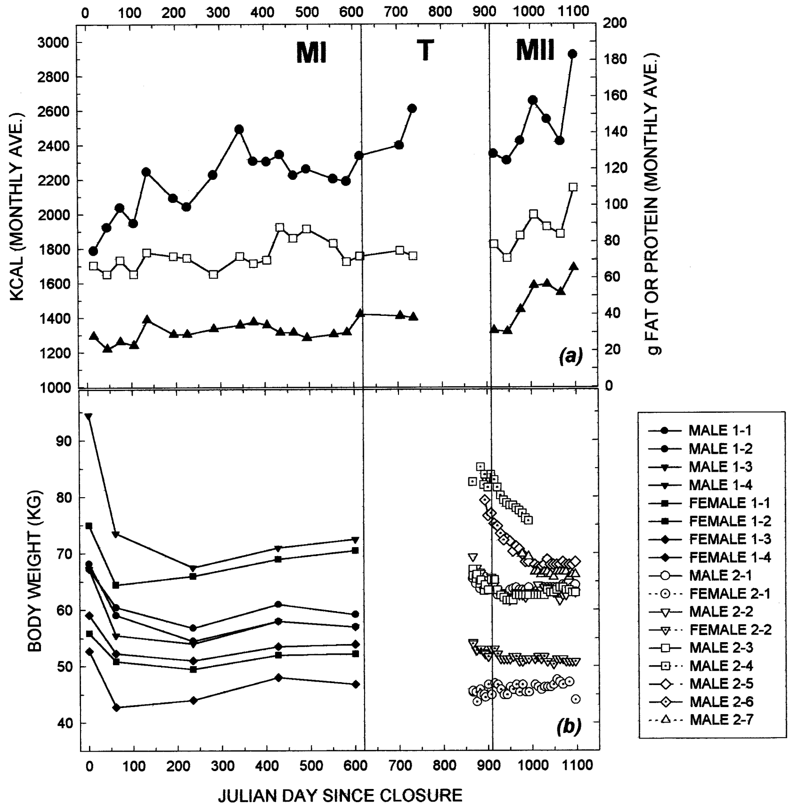

Fig. 6. Nutritional data for the IAB crews. (a) Nutrient data (plotted as monthly averages) for crews of Mission I and II obtained from the Biosphere 2 archival data base. (b) Weight loss of crews of Mission I and II; data obtained form Walford et al. (1996) and from the Biosphere 2 archival data base.

Another potential problem which was not assessed in either group of inhabitants was that of possible vitamin D deficiency. Since almost all ultraviolet light was prevented from entering Biosphere 2 due to adsorption of these wavelengths by the glass panels, skin synthesis of vitamin D would not take place. It is possible that intermittent vitamin D supplementation was sufficient to prevent deficiency of this vitamin, however, vitamin D levels were not measured. The cases presented above for vitamin intake emphasize the importance of careful monitoring of important metabolites to ensure adequate supply levels for long-term projects as well as the need for compositional analysis of foodstuffs grown under atypical conditions.

4.3. Integrated pest management for Mission I and II

Pest management was an integral part of optimizing crop production for both Missions, however, the rapid growth and establishment of two species, the crazy ant (*Paratrechina longicornis*) and the Australian cockroach *(Periplaneta australasiae*), according to available records, imposed a harsher consequence for Mission I than for Mission II (Silverstone and Nelson, 1996). Agricultural pests were not purposefully introduced into Biosphere 2 but rather gained entry during construction and initial planting of the Biosphere 2 flora. There were many species of harmful insect pests in the IAB that caused significant damage to crops particularly during Mission I when densities of some pests were dramatic, growing well beyond their natural densities due to favorable year-round environmental conditions, nutrient-rich soil and the absence of natural enemies. For example, the cockroach, not normally an agricultural pest, grazed heavily on a wide variety of crops, particularly ripe papaya, tomato, bananas, squash, eggplant flowers, new crop seedlings or transplants and leaves of the sweet potato and cabbage. As of June 1994, the ratio of insect and mite pests to natural enemies favored the pests by about 5:1, a situation that does not typically occur in nature or in agricultural settings (J. Litsinger, report on file, 1994). A summary of the pests present during Mission I are described in Silverstone and Nelson (1996). The yield reduction due to pests was dramatic; the broad mite and powdery mildew (the main plant diseases of the IAB recorded in October, 1993) destroyed entire plantings. As of early 1996, IAB insect pests included cockroaches, aphids, crazy ants, broadmites, spidermites, pill bugs, sow bugs, mealybugs, scale insects, leaf hoppers, white fly, flea beetle, banana weevil, psyllids, crickets, collembola, symphilids, and nematodes. Some natural enemies to these pests were present including spiders and introduced insects as described below. Pollinators remained absent since Mission I primarily due to predation by ants and cockroaches; introduced bee colonies did not survive probably also due to ant predation.

To combat insect pests a number of beneficial insects were introduced in the IAB as part of a biological insect control program before and during Missions I and II. They were (J. Litsinger, report on file, 1994): *Hippodamia convergens*—Coccinellid beetle (aphid predator), *Cryptolaemus montrouzieri*—lady beetle (mealybug predator), *Chrysoperla rufilabris*—green lacewing (aphid predator), *Orius tristicolor*—anthocorid minute pirate bug (thrips predator), *Lysiphlebus testaceipes*—braconid wasp (aphid parasite), *Leptomastix dactylopii*— encrytid wasp (mealybug parasite), *Metaphycus helvolus*—encrytid wasp (black scale parasite), *Nasomia vitripennis*—Pteromalid wasp (house fly parasite), *Diglyphus isaea*—eulophid wasp (leaf miner parasite), *Aphytis melinus*—aphelinid wasp (red scale parasite), *Encarsia formosa*—aphelinid wasp (white fly parasite), *Ambylyseius melinus*—phytoseiid mite (thrips predator), *Phytoseiulus persimilis*—Phytoseiid mite (spider mite predator), m*etaseiulus occidentalis*—phytoseiid mite (mite and thrips predator), *Neoseiulus californicus*—phytoseiid mite (spider mite predator) and *Amblyseius barkeri*—phytoseiid mite (spider and broadmite predator). Of the above introduced predators, however, few became established for a variety of reasons, including extinction of target pests, behavioral traits that were incompatible with a greenhouse environment and destruction by ant predation. However, the predatory mite, *Amblyseius cucumeris* was reared on a large scale and released in the IAB regularly during Mission II and was highly effective for broadmite control. Also introduced were 50 toads (*Bufo marinus*) and 40 Toke Geckos (*Gekko gecko*) for the control of the cockroach; both of these populations appear to have diminished significantly since introduction.

A large number of roaches were trapped using a glass jar. In addition, a 0.3-m tall aluminum sheet was implanted around the planting area for some plots to prevent the entry of roaches and mice onto the cropping area. Manual removal of aphids, mealy bugs and scale insects by washing with water, pruning and trimming proved to be highly effective.

Twenty different genera and species of plant parasitic nematodes were detected in Biosphere 2 soils during surveys completed in 1993 and 1995 (M. McClure, reports on file). A total of 70 sites were sampled in the IAB and orchard, nearly all of which had been sampled during the 1993 survey. One exotic nematode species, *Radopholus similis* (burrowing), was identified on a single banana plant. A new species, *Hemicycliophora biosphaera* n. spp., (Chitamber et al., 1997) was also found in the IAB soils. The nematode problem was actively managed during Mission II by planting areas of high nematode density with less susceptible crop species such as rice, corn, and sorghum. Also effective against nematode populations was the conversion of dry-land plots into wetland rice paddy plots.

4.4. Agricultural production

The cultivation of crops on a rotational basis proved to be of great benefit in agricultural pest mitigation along with the biological approaches discussed above. Cereals, vegetables and root and tuber crops were rotated with legumes in order to minimize the pest infestation while adding nitrogen to the soil. Intercropping methods of crop cultivation were also extensively employed. Cereal/cereal (e.g. upland rice with corn) or cereal/legume (e.g. sorghum with soybean, corn with beans or peanuts) based intercropping was practiced and was very successful during Mission II. The crop yield per unit area increased dramatically which ultimately increased the food intake and diversity. Crops with intrinsic insect repelling substances such as garlic and onions were also planted in alternate rows in some areas to repel and minimize the insect damage to the main crops.

It is likely that crop productivity could have been further improved by increased usage of transplantation rather than by direct seeding. IAB crops were planted by direct seeding (except for paddy rice) for both periods of closure, however, a number of problems emerged with this approach. Often, crops were over seeded resulting in poor stands due to reduced germination and damage to seedlings by pillbugs and cockroaches as well as by simultaneous emergence of weeds. Studies carried out in the analog greenhouse with IAB species using a transplantation approach demonstrated the potential for increased yield in part due to more efficient use of field plots resulting from the 2- to 3-week lag time between germination in trays and transplantation in the field plot.

4.5. Regeneration technologies

Regenerative technologies linked to the IAB were a combination of biological and physio-chemical approaches to treat waste water (urine and feces), to produce potable water and water for replenishment of the primary storage supply, to recycle plant inedible waste by composting and ingestion by animals, to supply and modulate the nutrient composition of the IAB soil, and to modify the composition of the atmosphere. The systems employed by the designers of Biosphere 2 were technologically simple and of much larger scale compared to analogous systems envisioned for space habitations or space travel consisting typically of a variety of highly engineered physio-chemical processes and microbially-based biological systems (e.g. Binot et al., 1994; Barta and Henninger, 1994; Strayer and Cook, 1995; Finger and Alzaraki, 1995). Results for regeneration of water, air and soil and recycling of biomass are discussed below.

4.6. Water quality

A nutrient supply system for the IAB was not specified for the periods of closure; nutrients supplied to the plants were derived exclusively from the irrigation water plus nutrients contained in mulch and animal manure (Silverstone and Nelson, 1996; Nelson et al., 1999). The salinity of the irrigation water was as high as 2000 mg l^{-1} total dissolved solids (TDS), as measured by conductivity, and contained up to 100 ppm nitrogen, measured as nitrate, by March of 1995. Nitrate concentrations of about 20 ppm and 40 ppm were measured in February of 1993 and March of 1994, respectively. The reason for the saline and nutrient-rich water was an ineffective water recycling system. Water for irrigation of the IAB, as discussed above, came from a variety of sources including the wastewater system (human and animal feces, urine and other waste streams), IAB sub-soil water, condensate produced from air handlers, indirect additions from the primary storage or a combination. The primary storage reservoir received untreated sub-soil drainage water primarily from the IAB and the wilderness biomes. In the case of the primary storage, the water quality declined over time due to ineffective methods for releasing fresh water from brine on a large scale, a process that clearly would have been necessary for long-term closure, that would have eliminated the need for RO

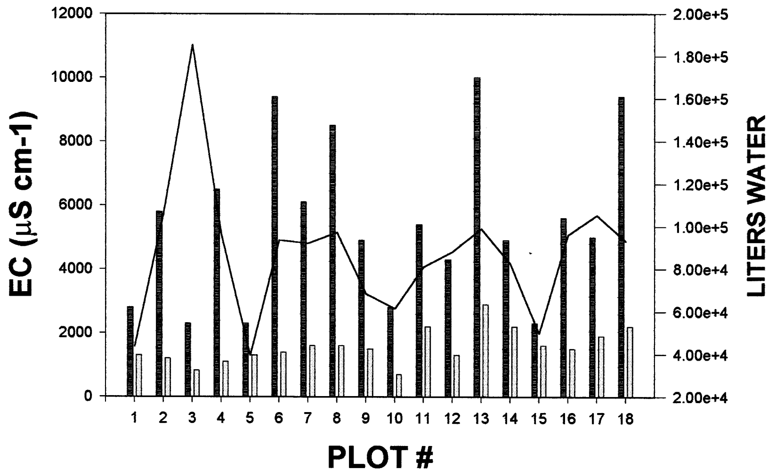

Fig. 7. Flush of IAB soils (indicated by plot # as shown in Fig. 1a) with treated well water to remove salts. Bars show initial (solid) and final (open) salinity (measured by electrical conductivity); solid line shows amount of water used for each plot.

machines and that would have provided ample high quality water for IAB irrigation. Indirect application of primary storage water and direct application of wastewater effluent delivered high concentrations of nutrients and salts to the IAB soils. Missions I and II irrigation water for the IAB was routinely saline and rich in nitrate and other nutrients, conditions which can affect plant productivity (Loomis and Connor, 1992). Subsequent to Mission II, a program of supplying relatively pure water collected from air handler condensate pans was implemented to assess the water production capacity of Biosphere 2 relative to the fresh water needs of the biota. Results from these periods of operation clearly indicated that during the summer months fresh water supply by condensation alone was not adequate; RO units were subsequently added, as discussed previously. The consequence of these changes was a dramatic increase in water quality delivered to the agricultural plants and the Biosphere 2 biota as a whole. The entire primary storage water supply was exchanged with fresh, treated well water during the summer of 1995. Current operating criteria specifies the total dissolved solids (TDS) set-point for the IAB irrigation water at 50 mg l^{-1}; higher values indicate contamination and an alarm condition is initiated.

Increased salinity and nutrients in the IAB irrigation water greatly influenced the inorganic composition of the IAB soils. The accumulation of salts could be seen on the surface of IAB soil in the form of a white crust. A soil salinity survey performed in 1995 clearly showed that most of the IAB plots were too saline for optimal plant growth. Reclamation of soil was effected by leaching with local well water of about 0.28 mS cm^{-1} salinity. The pre-leaching salinity level of soils, amount of water used for leaching and salinity level after leaching are shown in Fig. 7. The results clearly show that the soil salinity was effectively reduced by leaching with good quality water. Thus, in the context of a productive agricultural mesocosm, the soil quality was sustainable, but only with an adequate supply of fresh water produced by importation, as employed here, or by effective recycling of used water. Higher water quality during Missions I and II likely would have produced higher crop yields.

The effective processing of solid wastes, particularly human and animal feces, is crucial to a successful regenerative life support system. As described previously, the animal and human waste system was based on previous work by Wolverton (1980) employing plant and aquatic lagoons. Few data are available to assess the effectiveness of this system in reducing levels of organic and inorganic compounds (Silverstone and Nelson, 1996; Nelson et al., 1999). Given the observed levels of salinity in the IAB soils and the very high levels of inorganic nitrogen present in the primary storage reservoir it appears that the marsh system did not exert a significant level of control over these constituents resulting in increasing storage of inorganic nitrogen rather than effective recycling. Moreover, based on analyses of the potable water available for testing in 1994, it is unlikely that the wastewater system in conjunction with the water purification systems produced pathogen-free drinking water. An examination of the waste water effluent was undertaken in late 1994 under normal operating conditions of the waste water treatment system to assess water quality. The results of the study revealed the presence of protozoan parasites (*Giardia* and *Cryptosporidium*) as well as evidence of contamination by enteric viruses. High levels of coliforms and fecal coliforms were also detected. Since this effluent was routinely used to irrigate crops and subsequently routed to the primary storage tank the transmission of these pathogens to all water reservoirs was highly likely and thus posed a health hazard by water contact to those involved in agricultural and food processing activities. An examination of the potable water produced under normal operating conditions revealed the presence of the same enteric protozoan parasites and coliforms above the accepted safe levels for human consumption. The waste water treatment system and associated water purification systems were therefore not only ineffective in controlling nutrients and salts but evidently did not eliminate pathogens from the fecal waste stream. This is not surprising as the plant-based system utilized was not developed to produce microbiologically safe effluent for potable water or for crop irrigation; additional steps such as heat treatment are required to eliminate health hazards (Wolverton, 1980; Wolverton et al., 1983). The addition of a PeroxPure ozone/hydrogen perioxide treatment system was incorporated in the purification scheme to further sanitize the potable water and may have proved effective for Mission I inhabitants based on human health data presented by Walford et al. (1996) in which illness due to poor water quality was not reported.

4.7. The soil and ambient atmosphere

The trace gas records for the two periods of closure for the Biosphere 2 atmosphere have been discussed by Marino and Odum (1999). Here we address the atmospheric impacts of the IAB operation and management. The agricultural soils were enriched in organic carbon, as discussed previously, and thus were the largest source of CO_2 to the atmosphere. The loss of atmospheric oxygen from Biosphere 2 has also been documented (Severinghaus et al., 1994), although, as for CO_2, a quantitative treatment of the stoichiometry is not possible based on available data. However, it is clear that the addition of large amounts of organic carbon to the IAB soil was inconsistent with project goals since the quality of the air proved to be a life threatening and difficult ecosystem management problem being too low in O_2 (reduction from 21 to 14% by January 1993) and too high in CO_2 (as high as 5000 ppmv) (Walford et al., 1996). The high organic carbon content of the IAB soils increased the overall levels of CO_2 in the Biosphere 2 atmosphere year-round and had a negative impact on the coral reef due to acidification of the ocean water by the elevated CO_2. While the elevated CO_2 atmosphere enhanced the growth of agricultural plants (as well as of wilderness plants) the reduction in atmospheric O_2 was inconsistent with the goals of a life support system. The imbalance was due simply to consumption of O_2 by soil microbial activity (resulting in CO_2 which then was sequestered by the concrete) over the net production of O_2 by photosynthesis; the problem was worsened during the winter months when light levels were lowest. A reduction in the initial loading of soil organic carbon in the IAB soils would have diminished these problems. A chemical scrubber was employed to reduce the overall atmospheric CO_2 concentration. The byproduct of the scrubber (calcium carbonate ($CaCO_3$)) was removed during the summer of 1995 due to lack of effective technology for conversion to CO_2 (initially planned but not tested or implemented; Nelson et al., 1994), storage space constraints and health hazards.

While the soils of the IAB started with known additions of organic carbon, changes in soil organic carbon (SOC) content within the soil column through time were not well documented. A downward trend for SOC is noted based on analyses performed in 1993 (Franco-Vizcanzo et al., report on file), 1994 (E. Kelly, CSU; report on file) and 1995 (in house analyses; report on file). Using average values for combined plot and profile analyses, %SOC decreased from about 3.2% (0–12 cm; plots 1, 2, 7, 8, 12, 16, 17) to about 2.8% (0–10 cm; plots 1–18) 1994) to 1.88% (0–90 cm; single column profile of plot 10) as measured in 1993, 1994 and 1995, respectively. Thus, the high flux of soil CO_2 initially observed for the IAB soils has diminished since 1991, and, depending on environmental conditions and agricultural practices, will likely continue to diminish and thus lessen the associated problems discussed above. As a guide to production and consumption of the IAB soil trace gases (methane, nitrous oxide and carbon dioxide) under conditions similar to those for Mission I and II, samples were collected from different depths during transient closures in 1995 under variable CO_2 concentrations using a gas sampling probe at depth and analyzed for fixed gases by gas chromatography. Based on the results of these studies given in Table 4, the IAB soils were a major source of CO_2 to the atmosphere, a sink for CH_4 and primarily a source for N_2O (the largest N_2O flux was found in the Biosphere 2 rain forest during the periods sampled, followed by the IAB). The atmospheric levels of N_2O rose steadily throughout Mission I and II reaching as high as 100 ppmv, nearly 300 times higher than current ambient levels. In the absence of a simulated stratosphere and/or additional sinks for N_2O, the increasing trend for this gas was expectable.

Table 4
The range of flux of different gases in the IAB (μmol m^{-2} s^{-1})

	N_2O	CO_2	CH_4
Minimum	-1.13×10^{-3}	8.21×10^{-2}	-2.33×10^{-3}
Maximum	1.53×10^{-3}	6.82×10^{-1}	-1.71×10^{-4}

The soil bed reactor has been described as an effective device for purifying the air of problematic compounds resulting from outgassing and other sources (Nelson et al., 1994), however, no data are available to evaluate its effectiveness. During Mission II and thereafter the soil bed reactor was not used under closed conditions due to uncertainties in the performance of the system and possible health hazards such as increased particulates that might act as allergens, increased N_2O and CO_2, and possibly further reductions in atmospheric O_2. Thus, the SBR concept remains unproved at this scale. The apparent reduction in some atmospheric contaminants, such as volatile organic compounds, during Mission I could have been due to adsorption of certain compounds by the massive surface area of glass, metal, carpet and concrete or to atmospheric removal by rain and moisture. Plants are also known to remove a variety of organic compounds by introduction into stomatal pores (e.g. Porter, 1994).

5. Conclusions

As a food production facility to sustain human health the agricultural mesocosm of Biosphere 2 was very successful as a result of intensive farming practices, high biodiversity, high nutrient and CO_2 levels for growth and adequate water supply. However, as an integrated life-support facility, regeneration technologies that were in place during the first 3 years of operation were not effective in recycling water, solid and atmospheric excesses. The flaws in these systems could be easily overcome. Overall, the system was a remarkable success given the scale and requirements for operation. Stored waste products imply that regeneration, physio-chemical or bioregenerative, were not fully functional. Conversion of biomass to CO_2 was not needed over the time period examined due to the large buffer of available carbon in soils and the biota; the opposite problem of sequestering CO_2 was not adequately solved during either mission. Biosphere 2 accumulated very large amounts of stored waste in the primary storage facility as salts and other nutrients, stockpiles of wood as storage of carbon and very large amounts of $CaCO_3$ as stockpiles of removed CO_2, a clear indication that internal recycling was actually quite low, not complete or 100%, as indicated elsewhere (Nelson et al., 1994; Silverstone and Nelson, 1996). Major improvements needed for long-term, closed-system operation include: (1) a CO_2 control system that could sequester and release CO_2 upon demand (2) efficient evaporators or similar technology to recover fresh water from brine; (3) balancing of soil organic carbon content and associated soil CO_2 flux with production of O_2 by modifying soils and/or standing biomass; (4) an effective potable water treatment system; and (5) integrated control over nutrient flows between soil and water reservoirs. Presumably, these systems would have been upgraded over the envisioned 100-year life time of the project based on results from Missions I and II.

Although one of the original motivations for the Biosphere 2 project was to develop an application for habitation of the Mars surface (e.g. MacCallum et al., 1991) the Biosphere 2 IAB and associated systems shared little with prevailing approaches for life support systems in space which are conceptually based on attaining a balance between CO_2 and O_2 production and consumption. In this approach the need for re-supply of O_2 and exportation of CO_2 is eliminated (i.e. Controlled Ecological Life Support Systems (CELSS), e.g. Averner, 1989; Kliss et al., 1994; Binot et al., 1994; Gitelson and Okladnikov, 1994, 1996; and Advanced Life Support research (ALS), e.g. Barta and Henninger, 1996; Henninger et al., 1996). These approaches also relied on intensive monitoring of plant function (i.e. photosynthesis and respiration) and the use of models to maximize plant yield and control gas exchange (e.g. Wheeler et al., 1993; Volk et al., 1995). In the case of Biosphere 2, as illustrated above, external O_2 was supplied as a liquefied gas (three injections were made in 1993, two in 1994 and one in 1995) and internal CO_2 was removed by chemical scrubbing for both Missions I and II; no plant physiological data for agricultural plants, nor of IAB system wide cycling (carbon, water, nutrients) have been reported (cf. Nelson et al., 1994; Silverstone and Nelson, 1996). The primary differences were the use of natural soil versus hydroponic media for plant growth, scale (e.g. the air volume of the IAB is about 300 times that of the NASA Biomass Production Chamber; Wheeler, 1992), regenerative technologies and mission goals. The large scale and complex mechanical and biological nature of Biosphere 2 relative to CELSS facilities render many comparisons difficult; however, some insights are of potential relevance to space programs.

For example, the crop diversity achieved in Biosphere 2 demonstrated the health and aesthetic advantages of a soil system versus a strictly hydroponic one in which a small number of crops are grown, usually in monoculture. Inclusion of hydroponic systems, bioreactors and other solid waste processors in the IAB, as well as testing of solid matrix analogs to a lunar or Martian regolith (Nelson et al., 1994; Spomer, 1994; Ming and Henninger, 1994; Barta and Henninger, 1996), could be employed in the future in the context of related CELSS research. High CO_2 concentrations, ideally generated from soil organic matter or from a specified imbalance between total respiration and consumption in a closed ecological system, will be advantageous for crop productivity, especially for C_3 crops, many of which are high carbohydrate yielding. Higher photosynthetic rates imply higher O_2 production but loss of O_2 by CO_2 sequestration by concrete (see Severinghaus et al., 1994, for estimates of CO_2 uptake by concrete) or other O_2 sinks must be known. The IAB and perhaps habitat of Biosphere 2 could be viewed as an advanced, large-scale, soil-based, human-rated test facility for development of a combination of biological and physio-chemical life support technologies, much like the current closed system program of the Human Rated Test Facility, Johnson Space Center, under development (e.g. Henninger et al., 1996).

What did we learn about sustainability under varying environmental conditions? High crop and edible plant biodiversity was the single consistent factor in the design, planning, construction and operation of the IAB, clearly adding to the well being and overall nutrition of the crews and perhaps improved ecosystem function (Naeem et al., 1994). In the context of studies of sustainable agriculture, a future with high CO_2 and high nutrient availability could enhance the overall yields and biodiversity of C_3 based sustainable agricultural systems providing sufficient water is available. Unlike the concentrations of CO_2 in the Biosphere 2 atmosphere, the Earth's atmosphere is not likely to exceed 4000 ppmv over the coming decades (IPCC, 1996); however, it is clear that this system, with modification, could be used to study aspects of agricultural sustainability under simulated future conditions (CO_2, temperature, nutrients, etc.) providing a unique approach to the study of agriculture and global change research (e.g. Marino, 1997).

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