applied sciences

MDPI

Review

How IoT Can Integrate Biotechnological Approaches for City Applications—Review of Recent Advancements, Issues, and Perspectives

Pavel Gotovtsev

National Research Centre "Kurchatov Institute" Biotechnology and Bioenergy Department, 1 Akademika Kurchalova pl., 123182 Moscow, Russia; Gotovtsev_PM@nrcki.ru

Received: 13 April 2020; Accepted: 8 June 2020; Published: 9 June 2020

check for updates

Abstract: There are a number of significant changes taking place in modern city development and most of them are based on the number of recent technological progress. This paper provides a review and analysis of recent approaches of biotechnology that can find a place in today's cities and discusses how those technologies can be integrated into a city's Internet of Things (IoT). Firstly, several biotechnologies that focus on rain gardens, urban vertical farming systems, and city photobioreactors are discussed in the context of their integration in a city's IoT. The next possible application of biofuel cells to the sensor network's energy supply is discussed. It is shown that such devices can influence the low-power sensor network structure as an additional energy source for transmitters. This paper shows the possibility of bioelectrochemical biosensor applications, discusses self-powered biosensors, and shows that such a system can be widely applied to rainwater monitoring in rain gardens and green streets. Significant attention is paid to recent approaches in synthetic biology. Both cell-based biosensors and bioactuators with synthetic genetic circuits are discussed. The development of cell-based biosensors can significantly enhance the sensing possibilities of a city's IoT. We show the possible ways to develop cyber-physical systems (CPSs) with the systems mentioned above. Aspects of data handling for the discussed biotechnologies and the methods of intelligent systems, including those that are machine learning-based, applied to the IoT in a city are presented.

Keywords: Internet of Things; smart city; wireless sensor networks; biofuel cells; biosensors; biotechnology; synthetic biology; cyber-physical systems

1. Introduction

Internet of Things (IoT) became a major instrument that can significantly help modern cities to achieve their sustainable development goals [1–3]. Today, the IoT approach became a significant basis of the smart city [2] The active development of the IoT for cities goes alongside many other changes in the city itself. Modern cities have focused on such approaches as green architecture, new ways of planning, humancentric inclusive streets, energy efficiency, and many others [4–7]. Many of those approaches' biotechnologies are also becoming an influencer on the view of our future [8], including the view of the city [9]. The term "biotechnology" covers a number of different approaches in the many fields of application including medicine, pharmacy, agriculture, and environmental technologies, among others. Here, only a very small part of those approaches will be discussed. The main focus will be done on the application of living organisms for different city needs as a part of green infrastructure. This small part of many biotechnological approaches already today can give the possibility to produce food and treat air and water in the city, thus sufficiently influence the city's sustainable development. Both city biotechnology and city IoT develop almost in parallel at the same time and the necessity to provide research in the field of integration for those technological approaches is arising [1,8,10].

The most actively discussed biotechnological approaches for new production in the city are as follows:

- Higher plants as a major air treatment biosystem of cities: today there are several technologies such as rain gardens and green streets where higher plants also provide rainwater treatment [11,12];
- Urban vertical farming systems as a novel food-producing paradigm [13]: it is necessary to mention that such technology not only actively uses a high level of automation and robotization but has also become an effective carbon dioxide consumer that positively influences the city environment;
- Photobioreactors for microalgae production that can be a part of the architecture or for home air treatment [14,15]: phototrophic microorganisms have a higher rate of carbon dioxide fixation and can be the source of multiple useful substances that can be applied from biofuel and the production of biopolymers to fertilizers, food, and even the pharmacy industry [16]; additionally, it has been shown that phototrophic microorganisms can participate in rain and thaw water treatment [17], so photobioreactors can be part of a city's water system.

These technologies require additional operations to include them into a city's IoT. Their applications lead to new data generations and new control systems that operate with additional sensors that are needed to be added to the wireless sensor networks (WSNs) [10]. However, several recent advancements in the biotechnology can give rise to additional opportunities that enhance IoT capabilities in some fields. Some of those approaches are as follows:

- Energy harvesting technologies based on microbial fuel cells (MFCs) that are able to provide a power supply for IoT wireless sensors [18] that can operate consuming organic compounds from different wastewaters [19];
- Novel sensors for environmental monitoring based on biological sensing that can enhance chemical and biological sensing for IoT [10,20];
- New actuating systems with living cells integration [21] based on the synthetic biology approaches that give rise to the ability to develop a novel interface with biotechnological systems in the city [22].

Figure 1 illustrates these biotechnologies and their interactions with a city's IoT.

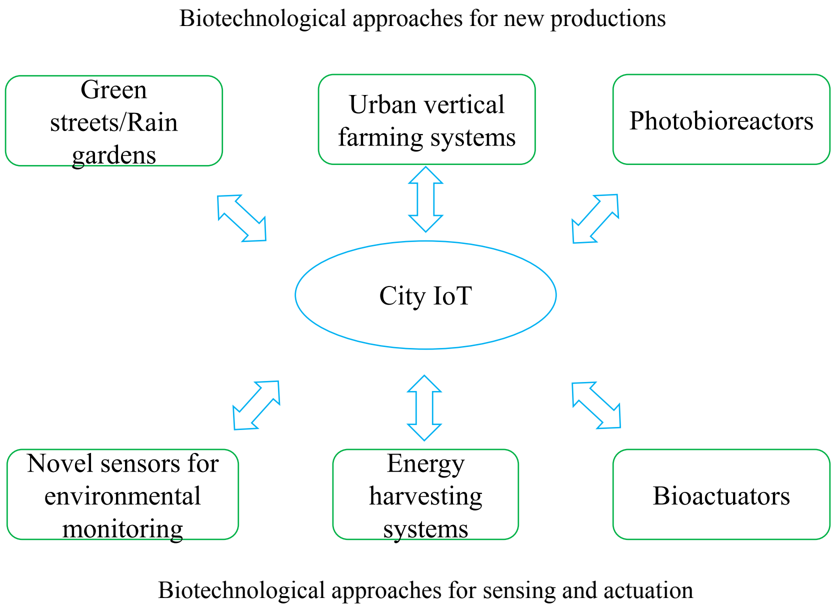

Figure 1. Interactions between several biotechnologies and a city's Internet of Things (IoT).

Thus, in this paper, the integration of both biotechnology groups in the IoT will be discussed with a focus on the new possibilities and issues that will arise for a city's IoT. Additionally, other approaches that can enhance IoT possibilities in some fields of application will be discussed.

Next, the paper will be organized as follows: firstly, biotechnological approaches (according to Figure 1) will be discussed, secondly, specific issues related to IoT technologies will be shown, and finally, the Discussion and Conclusion section will summarize the presented in the paper data.

2. Biotechnological Approaches for New Production

What makes the production of biotechnological systems integrated in a city's IoT differ from that in other technical fields? The main differences are as follows:

- Biological processes are relatively slower, especially compared with a technical system's speed of operation;
- Higher plants and phototrophic microorganisms are highly dependent on climate conditions that require both local climate conditions measurements and basic climate prediction data transfer [10].

The main or at least a significant stage of the process is inside living organisms, and in most cases, there is no opportunity to measure parameters of those processes on-line [22]. This can be partially solved by synthetic biology approaches [23] and will be discussed later.

An overview of specific issues related to the above-mentioned production biotechnologies shall be provided here. The application of higher plants is a well-known way to make a city sustainable. Many approaches now involve the use of higher plants in rain gardens and green street technologies that give rise to the ability to treat rainwater in city streets. The integration of such technology in the IoT via WSNs is described in [10]: an additional WSN is required to control both water flow and water quality. Additionally, some at least periodical usage of cameras from a municipal video control system can be useful to control the basic status of higher plants [24]. All this data via a wireless local area network (WLAN) should be transferred and then analyzed. It is necessary to mention that a WSN of such technologies generates the most useful data during rainfall and water flow, so it is possible to reduce the amount of data via the shutdown of sensors in the period between rainfalls.

Urban vertical farming approaches usually involves highly automated systems that focus on the fast production of fresh vegetables and berries [25,26]. In the context of smart cities, there are three significant parameters for the city's government: water consumption, wastewater discharge, and carbon dioxide consumption. Since most of these production farms are private, these systems have their own control and automation and are able to provide the necessary data exchange. Usually, vertical farming producers focus on the usage of artificial illumination, but some projects rely on usage and natural illumination [27]. In this case, climate data are significant for such projects, which leads to the additional data transfer from city climate parameter measurement systems via a data cloud.

Photobioreactors for phototrophic microorganisms production is a unique technology due to the higher speed of carbon dioxide fixation, in comparison with higher plants, and the possibility to treat and reuse rainwater for culture medium preparation or other technical needs [17]. Thus, photobioreactors can become a part of a city's infrastructure or at least a part of a building's infrastructure. The effective operation of photobioreactors requires a substantial amount of data from different WSNs, both in-house and from the streets [10]. This leads to the necessity of an integration photobioreactor control system with house and city data clouds for effective data handling. Receiving data from nearby systems can lead to the efficiency of this technology. For example, it is possible to use carbon dioxide measurements from the street, a number of people in a building, and indoor and outdoor temperatures, among others. Thus, those systems become a part of a city's IoT. Increasing the amount of data leads to new possibilities to enhance data analysis. There are a number of studies today focused on the use of modern intelligent methods to control the operation of photobioreactors and predict intracellular parameters of artificial neural networks (ANNs) [28,29].

Thus, issues that arise during the integration of such biotechnologies in a city's IoT can be summarized as follows:

- The additional WSNs and additional data generated by biotechnological systems;
- The additional usage of data, generated by other WSNs;
- Data generated by indoor photobioreactor sensors usually having a lower measurement frequency in comparison with, for example, street WSNs, and photobioreactor control systems not needing data with a higher frequency for its operation [10] (the same can be mentioned for other technologies under discussion);
- Additional long-time data storage, where "long-time" sometimes means years due to relatively slow biological processes and its climate and, respectively, season dependence (this is actual for all data related to system operation, including external data);
- The need for more precise process monitoring methods such as ANNs;
- The need for additional wireless sensors for an additional power supply that is more actual for green streets and rain gardens, where sensors sometimes should be placed in unusual places such as drainage lines.

Next, how biotechnology can give rise to new possibilities in a city's IoT both to solve the issues mentioned above and to enhance its capabilities will be discussed.

3. Biotechnological Approaches for Sensing and Actuation

3.1. Biotechnology for Energy Harvesting

Installing a WSN or a network of Wi-Fi transmitters in streets and gardens requires solving the issue of power supply. Many approaches can be used, and one of them is based on the application of MFCs [18]. MFCs are able to harvest energy by consuming organic compounds (substrate) in rainwater or water in soils to generate electricity [30,31]. In such biofuel cells (BFCs), microorganisms are used as biocatalysts. For environmental use, MFCs with microbial communities as biocatalysts can provide more stability and reliability during long-time operation [32,33]. Another interesting approach is the integration of MFCs with higher plant or phototrophic microorganisms—phototrophic microbial fuel cells (PMFCs) [34,35]. Depending on the plants and microorganisms, a cathode or anode can be integrated with a plant. PMFCs can be a more stable and reliable energy source in comparison with MFCs under the same conditions, but further studies are needed [34,36]. Streets and parks can be a good option for further PMFC testing.

The MFC and PMFC power outputs strongly depend on the materials of the electrodes, the concentration of organic compounds, the bacterial biofilm in the electrode, and many other factors [32,37]. It is possible to await an unstable generation around several mW·cm-2 (mW per electrode surface) at achievable voltage values such as 0.5 V [18]. An overview of some MFCs and PMFCs is shown in Table 1, where it is shown that different types of MFC can demonstrate very close power output. In all MFCs, power generation strongly relates to the concentration of consumable organic compounds, the number of active cells on the electrode, and the speed of the biochemical reactions that take place in this system. Therefore, power output cannot be unlimited with the increase in the concentration of organic compounds [38]. Regarding unstable energy generation, it is necessary to use a power control system with energy storage in the battery. Therefore, MFC and PMFC applications require such a system for an effective sensor power supply [18,39]. This leads to the development of combined power sources that can include MFCs and, for example, small solar cells.

Table 1. Characteristics of several microbial fuel cells (MFCs) and phototrophic microbial fuel cells (PMFCs). Power output data given in the same scale as in the siting papers.

№	Type and Overview	Biocatalysts	Power Output	Reference
1	MFC with microfluidic system of chambers	*Geobactor*-enriched mixed bacterial biofilm	4.7 μW/cm^2	[40]
2	MFC with different strains of yeasts. Glucose and xylose were used in substrates	Several strains of yeasts. Most promising results shown with *Kluyveromyces marxianus*	2.5–3.32 mW with xylose. and up to 4.35–6.48 mW with glucose	[41]
3	MFC with modified by poly (3.4-ethylenedioxythiophene) anode	*Shewanella loihica*	140 mW/m^2	[42]
4	PMFC with different waste waters as substrate.	Microbial community from sewage sludge as biocatalysts in anode. *Chlorella vulgaris* in cathode chamber.	From 23.17 to 327.67 mW/m^2 during 32 days of stable operation.	[43]
5	Paper-based MFC operating under continuous flow conditions	*Shewanella oneidensis*	25 W/m^3 (25 W per cubic meter of both MFC chambers)	[44]
6	Microscale MFC	*Geobacteraceae* -enriched mixed bacterial biofilm	83 μW/cm^2 and 3300 μW/cm^3 respectively	[45]
7	Membranelles MFC from cheap materials. Applicable for applications in wastewater.	*Gluconobacter Oxydans*	1.43 μW/cm^2	[19]
8	PMFC for waste water treatment and cyanobacteria production	Anode–microbial community sewage sludge; cathode-*Scenedesmus acutus*	Up to 5.3 mW/m^2 and 400 mW/m^3, respectively.	[46]
9	PMFC for kitchen waste water treatment with phototrophic microorganisms in cathode chamber	Anode–microbial community from waste water.Cathode-*Synechococcus sp.*	Approximately 41.5 mW/m^2	[47]
10	Large-scale MFC (85 L volume)	Microbial community	0.101 W/m^2 and 0.74 W/m^3, respectively	[47]
11	MFC with air cathode and capability to total nitrogen removal	Microbial community with denitrificators	Approximately 1250 mW/m^2	[48]
12	MFC with cylindrical construction	*Pseudomonas aeruginosa*	Up to 3322 mW/m^2	[49]
13	PMFC with higher plants in different environmental conditions	Microbial community-based cathode	From 0.3 to 10.13 mW/m^2 under the different environmental conditions	[50]
14	PMFC with higher plants for IoT sensor power supply	Microbial community-based cathode	3.5 mW/cm^2 per single plant	[51]

Here, the possibilities of application of biofuel cells for low-power sensor networks in rain gardens and green streets will be discussed. MFCs can be both a main and additional power source for increasing the time between battery changes [10]. Experiments such as [18] show the possibility of using MFCs for powering sensors and providing periodical data transmission via low-power Bluetooth radio. It is necessary to mention that the MFCs used in this study can be improved via new electrode materials and more effective cell immobilization methods [33,52,53]. Thus, microbial MFCs can be used as a power source for sensor networks. This leads to the possible application of MFCs as local power sources for low-power wide-area networks (LPWANs) installed in streets and parks. A LPWAN is a very effective approach for installing energy-efficient sensor networks in smart cities [54,55]. Figure 2 illustrates possible variants of microbial MFC applications for an LPWAN component power supply. The most obvious method is applying MFCs to increase the operation live-time of wireless sensors (Figure 2A). To decrease the cost of the sensor network, MFCs can be used for the most significant sensors or for sensors whose operation stability is critical. MFCs can be used for a sensor's transmitter power supply to increase the distance of transmission (Figure 2B) or the amount of information (Figure 2C). Additionally, a combination of those variants can be used with respect to each project feature.

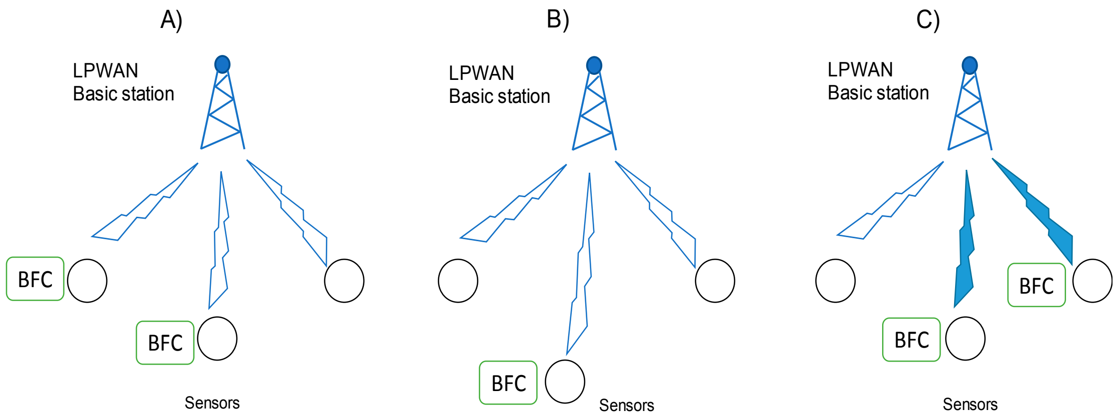

Figure 2. Variants of microbial fuel cell (MFC) applications for low-power wide-area networks (LPWANs). (**A**) MFCs used as a power supply for wireless sensors to increase the time of operation; (**B**) MFCs as an additional power source for sensor transmitters to increase the distance of transmission; (**C**) MFCs as an additional power source to increase the amount of data from sensors.

An interesting application of the variant presented in Figure 2C can be found in green streets or rain gardens. As mentioned in [10], such systems can use several sensors for water flow and quality control. Those sensors should operate during water flow, so there is no need to use it actively in dry periods. MFCs can also effectively operate when water flow with organic compounds has taken place. Therefore, in this case, MFCs can be used as a special power booster to supply sensors and increase the frequency of data transmission. Additionally, green streets and rain gardens can be a good place for PMFC testing and applications. Thus, it is possible to develop a bioenergy supply sensor network for green streets or rain gardens.

An LPWAN's basic station usually requires energy equal to tenths of W. Theoretically, it is possible to build MFCs that can generate such a power output in the case of the availability of organic substrate. Solving this issue and finding specific environmental conditions is the goal for further research.

MFCs can be applied for the power supply of the low-power wireless transmitter. Low-power Bluetooth transmitters usually require less than 1 W of energy that can be effectively generated by MFCs. This means that MFC-supplied transmitters can be used for heterogeneous wireless networks. Such networks that include different transmitters have become very popular for wide spaces and IoT applications [56,57].

Finally, it is necessary here to mention several issues related to MFCs in the applications mentioned above:

- Unstable generation related to substrate availability, which leads to a necessary power control system and battery;
- The possibility of microorganism death for several reasons that require additional research on long-time BFC operation in natural environments [33];
- Low power output that leads to the necessity of its combination with other energy-harvesting technologies.

Nevertheless, there is research focused on all these issues, and MFCs are still a promising energy harvesting technology for low-power devices for a city's IoT.

3.2. Biosensors for Environmental Monitoring

"Biosensors" usually refer to sensors with biological sensing parts, operating in cells and organisms to measure biochemical substances. In environmental monitoring, biosensors have become a subject of interest for many researchers due to the following advantages:

- Frequent low power consumption [52], including self-powered sensors [58,59];
- The ability to measure parameters that are difficult to measure in place and on-line and that are usually measured by laboratory methods [60];
- The possibility of multiparametric measurements using one cell-based biosensor [61,62].

All of the discussed biosensors can be divided into the three big groups:

- Bioelectrochemical sensors with enzymes or cells as the sensing part that generate an analog electrical signal;
- Biomolecular (DNA, antibody, and enzyme) sensors that require an optical detector;
- Cell-based sensors with genetically engineered cells that are able to provide multiparametric measurements and/or some intracellular processing [63].

Bioelectrochemical sensors are the most widely used because they have an analog electrical signal (current or voltage) that is easy to handle and process, in almost the same way as in the case of classical electrochemical sensors. The sensing part is the cell or enzyme and provides a bioelectrochemical reaction that in its turn generates output. There are a number of approaches to using such biosensors for measuring toxic pollutants, heavy metals, ammonium, and bioorganic compounds [30,64–66]. In many cases, such sensors are combined with BFCs because the measuring process involves the generation of electrical currents as a result of a bioelectrochemical reaction catalyzed by an enzyme or cell [64]. This approach leads to the possible application of such self-powered biosensors in rain gardens; they are able to start their operation with the start of measurements, so they can be self-activated during water flow appearance. Summarizing, it is necessary to say that bioelectrochemical sensors are an extremely promising field of research to enhance the sensing capability of a city's IoT.

Biomolecular (DNA, antibody, and enzyme) sensors are mostly used for laboratory measurements [67]. The fast development of optical biosensing [68] and related technology might give rise to opportunities for future applications of such biosensors outside the laboratory.

The fast development of synthetic biology in recent decades has yielded advancements that can lead to IoT applications. In the following, approaches that are able to expand sensing possibilities will be discussed.

The development of multiparametric cell-based biosensors is a synthetic biology achievement. One interesting approach to develop it is based on the integration of simple Boolean logic gates that synthetic biology can provide for IoT applications [22,69]. The logic process, in this case, provided through genetic logic circuits, is based on the variation of synthesis of active elements such as repressors and actuators that influence the biosynthesis processes from another part of the DNA [63]. It has been shown that tenths of logic gates can be applied in the cell [62,70]. Providing such computations in the cell can provide the following advantages for whole-cell biosensors:

- One cell can be made a multiparametric biosensor with an indication via fluorescent protein biosynthesis [63,71];
- Output can vary depending on the input parameters;
- A system can be developed with cells with different functions that can contact each other via chemical signals [22,70] and generate more readable and informative output [72].

Thus, it is possible to develop a multisensory array from several groups of cells that can provide not only sensing but also preliminary data analysis. The main advantage of such an approach is that microorganisms can support their operation via their metabolism, and the energy consumption of whole sensor systems begins from detection signals from cells. Figure 3 schematically illustrates such a sensor system. Cell group A provides sensing and a preliminary analysis of the measured signals and generates a chemical output that can be analyzed by the other groups of cells (cell groups B and C in Figure 3) for continuous processing and output generation.

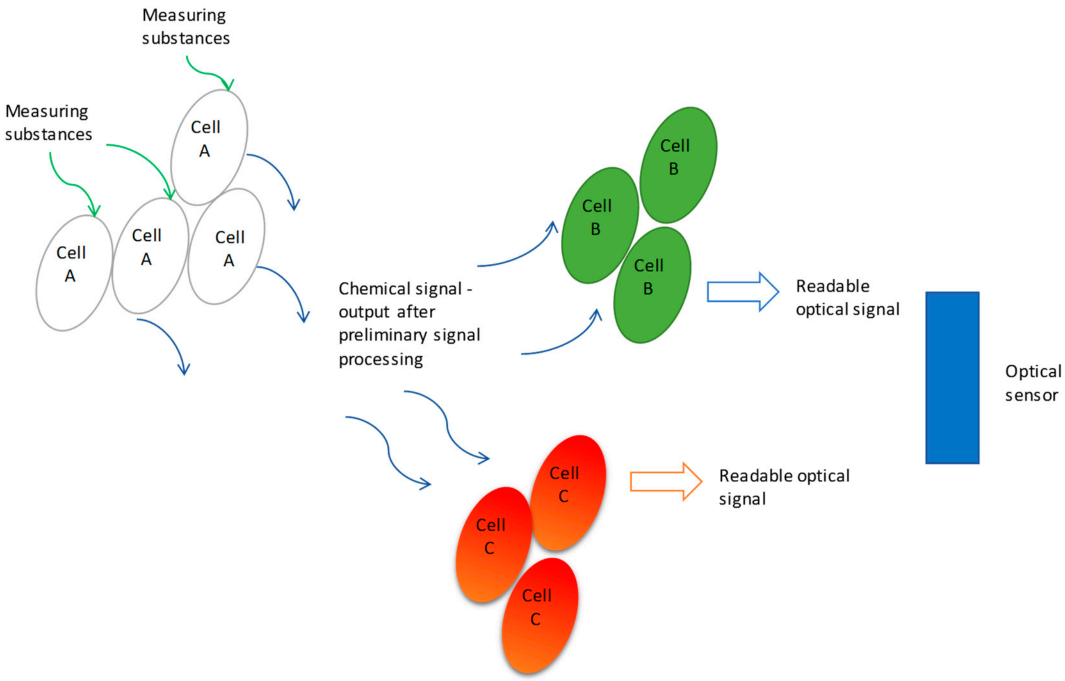

Figure 3. Multi-sensor cell systems with several stages of data processing.

Next, some limitations and issues related to such sensor systems are discussed from the perspective of their possible application for a city's IoT:

- There can be a long time between measuring substances arising, and response signals can result because of the need to provide many biosynthetic processes inside the cell, and this time can be equal to several hours [23];
- There is a possibility of situations that lead to massive cell deaths;
- There is intracellular resource competition—a lack of usable substrates that can prevent most of what is necessary for signal analysis biosynthesis and genetic logic circuit operation [73];
- There is less sensitivity in comparison with nanosized sensors or some electrochemical sensors;
- Difficulties occur with cell growth control and stabilization in an open environment, so the necessity of cell immobilization arises;
- Many cells influence the value of optical output, so an additional analysis of the measured data is needed to obtain reliable results;
- The long-time stable operation of such sensors in an open environment requires additional research;
- Legislation issues arise concerning the application of genetically modified organisms in an open environment, which also leads to the immobilization of cells and additional technical systems for preventing cell leakage [74].

Thus, some of those issues can be overcome using immobilization techniques [75] and using membranes between cells and the environment. Additionally, many studies have focused on the development of the modularity of engineered genetic circuits by adding retroactivity and feedback controllers to insulate modules [73,76,77].

The genetic logic circuit approach is not the only way to provide analysis using intracellular processes. Many studies have focused on analog synthetic biology [78], which has parallels between analog electric circuits and intracellular processing. This approach has many advantages such as the ability to develop hybrid systems with low-power electronics [79] and to develop new analog processes for measured data analysis; for example, some analogs of electrical circuit components have been developed [80]. It is also necessary to mention that the development of low-power cytomorphic electronics for biointerfaces is a promising tool for biosensors in IoT development [78,79].

Next, attention will be paid to the approaches based on synthetic and modified proteins. The development of synthetic and modified proteins is an issue because there are multiple variants of ways in which protein can make its conformation, but this approach can yield new, more stable, and effective enzymes [81,82] that can be used as the sensing element of biosensors. Protein systems can be used for intracellular computation [83]; for example, based on enzyme-catalyzed reactions, logic gates can be simulated [84]. A recent paper demonstrated the de novo design of protein logic gates that effectively operate in a leaving cell [85]. Protein logic gates are able to provide operations faster in comparison with genetic logic networks due to the possibility of the preliminary biosynthesis of several proteins that participate in the reactions.

Finally, the interface between cells and our common electronic systems will be discussed. As shown in Figure 3, mainly fluorescent protein biosynthesis is used as an output signal [72]. This indicates the necessity of optical sensing systems. However, this does not mean that this is the only way this interface can be built. It is possible to use chemical signal–small chemical substances that can be easily detected and transferred to the electrical signal. It can be a system with a unique small biomolecule and specific sensor molecule immobilized on the bioelectrochemical sensor. However, there are many issues that should be solved for the use of received signals as analogs of measured chemicals, including the number of nonlinear processes inside cells and diffusion processes in the environment. A possible way to solve those issues is to use machine learning approaches [86] and/or mathematical models of intracellular processes. Using digital twins, simulations in parallel with the real process, can be an effective way to enhance IoT WSNs [87] and especially cell biosensor data processing, but whole cell models are difficult to design and require substantial computational power [88]. One promising approach using analog electrical circuits for modeling signal processing related to reactions in the cell has been found. This approach shows efficiency in the modeling of intracellular processes [89,90].

3.3. Bioactuators

A modern IoT is comprised not only of different sensor networks but also of a number of actuating systems, such as climate control systems in buildings and different kinds of smart electronics—even traffic control lights [91,92]. These systems operate based on sensor data. Here, we will discuss what actuating systems for IoT can arise thanks to recent approaches in biotechnology.

Higher plants in rain gardens and green streets have already been discussed as a technology that can be applied in a city with IoT, which leads to additional WSN installation and thus to additional data generation. All sensors in such systems are focused on the control of rainwater flow and its chemical parameters [10]. Higher plant status is controlled via common street cameras. Technologies developed for smart farms can be used to support these plants, and additional sensing for plant states and delivery require fertilizers and nutrients by drones in case of some plant-based issues regarding the sensor data [93]. Such feedback requires additional resources, energy, sensors, more data to handle, etc. Energy-effective approaches based on the properties of biological systems will be discussed next.

Genetic logic circuits can be used not only for cell sensors. Using this approach, it is possible to provide different types of output from cells, connecting input and output via the logic circuit. This output can be, for example, biopolymeric molecules that have some perspective properties as materials or proteins that tie heavy metals in the water. Cells can be successfully integrated into materials for 3D-printed system and generate a predictable output [94–97]; thus, hybrid living materials (HLMs) [98], and biologically derived structural materials [21], were designed. HLMs and their analogs are a very promising approach that can change a material's properties with the response to inputs from an environment, natural or artificial, generated based on sensor data from a nearby WSN. In the first case, there is a closed biological system that can signal its state via the synthesis of fluorescent protein coupling with the biosynthesis of the necessary product. In the second case, it is possible to add small signaling molecules basing on the data from a WSN and start biosynthesis in the cells. Thus, novel materials with both sensing and actuating capabilities can be developed [99]. Table 2 shows several possible HLM applications.

Table 2. Possible applications of hybrid living materials (HLMs).

Possible Application	HLM Function	Input and Output Signal
Functional polymeric materials with sensing capabilities	Sensing of hazardous chemicals in the environment	Input—hazardous chemical in the environment Output—optical signal
Adaptive polymeric materials	Synthesis of biopolymers to change properties of material. It can be synthesis of degradable polymers on the non-degradable matrix for temporary effect	Input—additional chemicals or environmental factor. Output—optical signal
On/Off self-healing for polymeric materials	Synthesis of biopolymers for self-healing controlling external signals	Input—additional chemicals Output—optical signal
Reinforcement for concrete under the natural environment influence	Synthesis of biopolymers or participation in biocrystallization processes	It can be a constant process or as a reaction on the humidity changes around cells Output–optical signal or chemical signal

An interesting approach was presented in [100]. Using spores of the genetic engineering bacteria *Bacillus subtilis*, which can produce spores that cannot form a living cell, as an actuator that changes its volume under the influence of humidity, the authors designed different macrosystems, such as artificial humidity-driven muscles and rotary engines. This approach can lead to a humidity-driven system for building ventilation. As in the case of HLMs, this system can operate independently, generating only a signal with information on whether or not to operate it.

4. Integration into IoT

4.1. Data Handling Challenges

As in the case of any additional technology in a city's IoT, for the biotechnological approaches mentioned above, new data challenges and data handling specifics are arising. Regarding production biotechnologies, as mentioned above, the speed of biological processes is slow in comparison with most of a city's technical systems. A lower speed of processes leads to a lower frequency of measurements by sensors in the system and a lower amount of data to transmit [10]. However, this does not mean that it needs a lower amount of data to store. For green streets/rain gardens and on-street photobioreactors, unique features that influence the amount of data that need to be handled, and the data handling process in total, are as follows:

- Climate influence. Biotechnologies interact with the environment and are influenced by local weather changes. Day-to-day weather irreproducibility in different years leads to the need to collect data throughout many years in case machine learning systems are developed to control and predict the operation of the production biotechnologies under discussion;
- Heterogenic data by means of different frequencies of measurements by sensors, using those systems. This data should be processed and analyzed together, especially in the case of applications of predictive models;
- Video cameras that can be applied to control higher plants [24] in rain gardens lead to the necessity of somehow processing data from them for long-term storage;
- Periodic operation of some sensors that control parameters of the rain waterflow. There is no need to use them when there is no water in the system. This also influences the data heterogeneity

because rain does not happen on the same day and the same time with the same efficiency every year.

Thus, integration into IoT rain gardens/green streets and photobioreactors generates heterogenic data, including data from video cameras that need to be stored for years. This leads to issues with data analysis, especially when there is less heterogenic data from widely used city WSN sensors [10].

In vertical farming installations, as mentioned earlier, there are more closed systems in terms of data exchange with a city's IoT environment [13,87,101,102]. However, it is necessary to control the input and output resource streams, such as heat, water, wastewater, and electricity, and to provide quantification of the amount of carbon dioxide fixation during operation. All this data helps to effectively control a city's demand for water and energy and to calculate the impact of city farming on the city's carbon footprint. This yields additional data flow that, in comparison with previously mentioned production biotechnologies, is much less heterogenic.

Figure 4 illustrates the data flow between photobioreactors, vertical farming systems, and building control systems. In this figure, vertical farming systems have a high level of automation and operate only under artificial illumination in a controlled environment. Data from the sensors of both systems goes to its control systems. As mentioned earlier, data exchange between biotechnology control systems and building control systems is much less frequent. Data from a city data cloud can be transferred both via a building control system and directly in a photobioreactor control system. It is necessary to mention that it should be data from the same source in the case of direct data transfer. Additionally, collecting all data concerning photobioreactor operation in a city could lead to the development of new deep learning based-systems to control those systems.

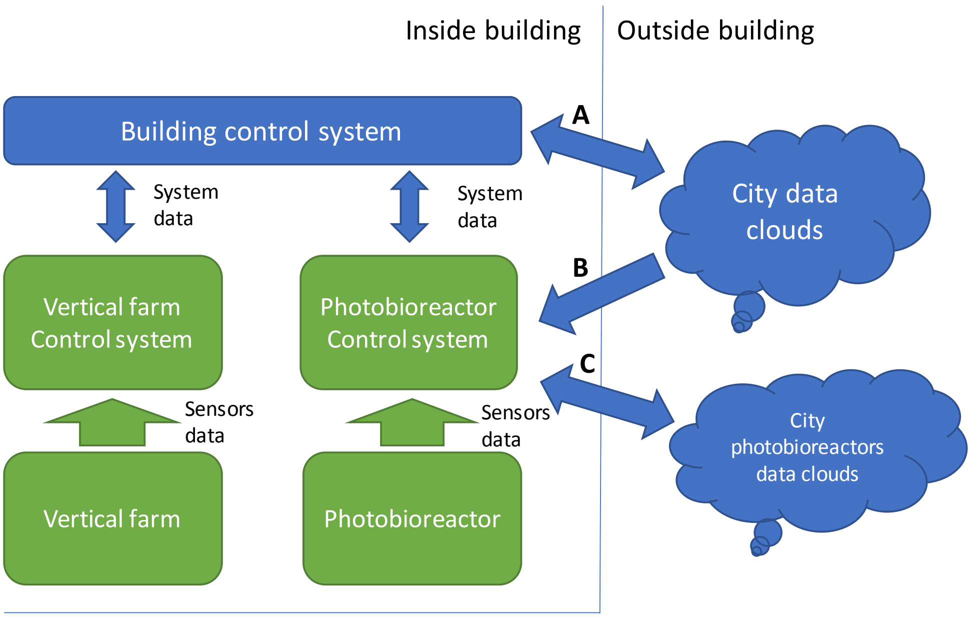

Figure 4. Data flows between photobioreactors, vertical farming systems, and building control systems. (**A**) City data transfer to a photobioreactor control system through a data exchange with a building control system; (**B**) data transfer directly to a photobioreactor control system; (**C**) data exchange between a photobioreactor control system and data clouds containing data from all photobioreactors in the city.

Can synthetic biology influence data handling in a city's IoT with the biotechnology mentioned above? Living matter is able to provide effective data storage and operation inside the cell using DNA and transcription/translation mechanisms. There are already existing approaches to the storage of non-biological data in DNA [103–106]. The ability to encapsulate DNA molecules in silica beads, which in turn can be put in polymeric systems for long-time storage, has already been shown [105]. Due to the long time and higher cost of DNA synthesis and the subsequent sequencing to extract the data, this approach today can be applied for long-time storage goals, addressing this data no more than a couple of times per year. One application for such data storage can be steganography [105].

More interesting for future IoT applications is the automatization of digital data transfer to functional biologics starting from DNA [107]. Many synthetic biology applications widely use standardization and unification approaches in, e.g., engineering disciplines, i.e., those based on standard biological parts [108,109], synthetic biology open language [63], and automated project development software such as CelloCad [69]. Such developments have led to engineering methods whereby biological systems are built based even on bacteria or yeast. This has led to the possibility of organizing the fast development and production of cell-based biosensors combining the biologics production and modern 3D-printing technologies mentioned above [110,111]. Such development technologies require additional effective data analysis techniques, which will be discussed next.

4.2. Intelligent Systems and Biological Objects in the IoT

Different intelligent methods, mostly from the machine learning field, are widely used in biotechnology today [86,112,113]. They have applications in genetics [112], protein structure prediction [114], and metabolic pathway dynamics prediction [115]. These approaches have been widely applied in the development of different types of biosensors, mentioned above. Here, it is necessary to mention a deep learning approach for the lab-of-origin prediction of engineered DNA [113]. Such a method can also be applied for cell-based biosensors quality control.

Here, the place of intelligent systems in the production biotechnologies mentioned above and how such systems can help with their integration with a city's IoT are discussed. The most obvious application is data from video camera analysis together with data from sensors to control the state of higher plants and the efficiency of rainwater treatment. Such systems based on machine learning approaches are already widely used for smart greenhouses, including vertical farming systems [87,102,116]. Therefore, the development of machine learning approaches for rain gardens and green streets can be an effective way to control such systems.

Machine learning approaches have already been applied to process control [117], the prediction of biomass growth [28,29,118], and the production of useful biochemicals [119]. Applications in laboratory and industrial photobioreactors have been shown. In the case of city photobioreactors, additional influencing variables should be taken into account. Main locations for the installation of such systems include building facades and roofs [120], the interior of big buildings such as railway stations, and shopping malls in places with natural illumination, similar to other green systems with plants [121]. This leads to the influence of climate conditions inside a building on biomass growth, so additional data from in-house sensors are required. The operation of a photobioreactor in turn influences the conditions in the building. Carbon dioxide consumption should be additional criteria for maximization since this is a main reason for photobioreactor applications in cities.

4.3. Cyber-Physical Systems

A city's IoT today is often associated with cyber-physical system (CPS) applications [87,122]. Many mentioned approaches can be described as CPSs, including automated vertical farms, which are widely described in the scientific literature [26,87]. In this section, CPSs related to biosensor and bioactuator approaches are the main focus.

As shown earlier, biotechnology provides the ability to make sensor systems more autonomous in terms of self-activation and preliminary data analysis. In some cases, such systems require unique methods of handling. Regarding self-powering MFC-based biosensors, MFCs, used for a biosensor's power supply in rain gardens and green streets, can start their operation and then water flow with organic impurities will arise in the system. After the rain ends, the water flow disappears and the MFCs shut down. The time of system operation can be controlled by using an additional battery to collect energy and select the power output of the MFCs [18]. Thus, the bioenergy source-based regulation of sensor operation can be realized, so more distributed and more autonomous WSNs can be developed.

As shown in Figure 3, multi-sensor cell systems with several stages of data processing include multiple stages in genetic logic circuits and cell-to-cell communications. As mentioned, there are many issues related to the application of those sensor systems that need to be solved. The development of digital twins of such sensors can be one of the ways of solving some of the mentioned issues. HLM operation also requires some predictability when operating in an open environment. HLM users should be sure that, as a result of the work, they receive necessary results without any negative consequences. Moreover, the predictability of synthetic biology-based biosystems is one of the key issues that make legislation regarding its application less prohibitive [74,123,124]. The prediction operation of those systems faces difficulties related to the complexity of the intracellular process [125]. Thus, to decrease the complexity of model approaches, the requirements of simulation processes need to be highlighted. It was mentioned earlier that simulations using analog electrical circuits can help to solve these issues. Thus, how this system with digital twins can operate will be discussed here, where on/off self-healing for polymeric materials will be used as an example (Table 2). Figure 5 illustrates an example of an HLM application. There are two WSNs applied in this system: one is a WSN for environmental control, and the other is much smaller for HLM control and includes optical sensors. When a signal from the environment (for example, chemical) arises, cells begin biopolymer production and fluorescent protein biosynthesis as a signal that they can start operating. This is a signal from cells, and it changes in HLM due to arising biopolymers that are registered by the HLM WSN. Digital twins provide simulations of HLM processes, are based on measurements from WSN data, and result in simulation control action development. Control action can consist of adding biochemicals that influence HLM operation. Thus, additional WSNs to control HLM are necessary to provide measurements and receive data that will be compared with digital twin simulations results.

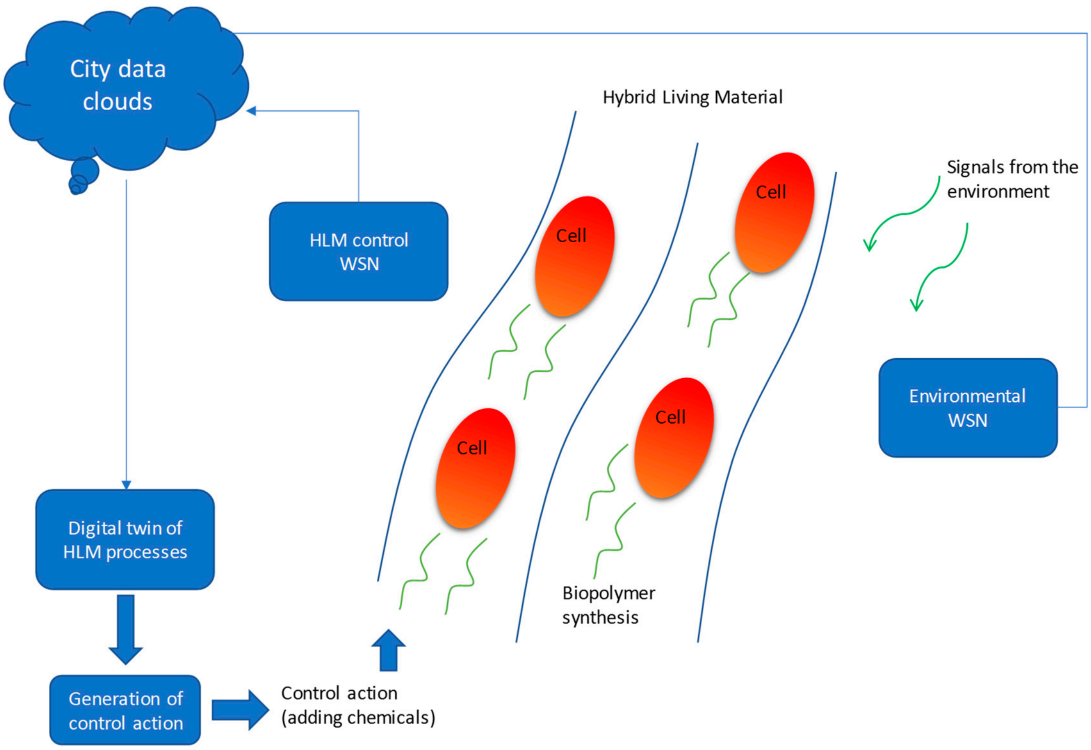

Figure 5. Cyber-physical system (CPS) with hybrid living material (HLM) and two wireless sensor networks (WSNs) for process control.

5. Discussion and Conclusions

The presented review shows that several modern approaches in biotechnology and synthetic biology can find applications in today's IoT-driven smart city concepts. Moreover, new possibilities in biosensor and bioactuator developments and applications are arising. Thus, further application ideas have arisen with the appearance of results of new studies. However, all technologies have issues together with advantages. The issues that pertain to most of the biological systems discussed can be summarized as follows:

- There are issues related to the complexity of biological systems. As shown in this paper, the biological object is sometimes a small element of the system, such as cells in cell-based biosensors, but processes inside this small cell are very complex and are influenced by many factors, which should be taken into account in the development of such systems;
- There is an environment dependence. Biological systems live in a narrow range of climate parameters, and that can be narrower for some species than common weather changes during the year;
- Interface issues persist. It is difficult to receive data concerning processes that take place inside the objects without their destruction. Synthetic biology yields some approaches that can help solve this issue, but its application requires additional sensors (optical, biochemical, etc.);
- There are legislation issues related to genetically engineered organisms, and overcoming these issues can provide a demonstration of the behavior predictability of systems with such organisms.

All advantages that can be enjoyed by biotechnology applications for a city are difficult to identify because some of the above-mentioned technologies are still at the laboratory research level. Nevertheless, based on the presented data in this paper, some advantages can be defined:

- Efficient data exchange between photobioreactors or urban vertical farming systems and a city's data centers can help to utilize data from all such installations for predictive mathematical models' development;
- Biofuel cells are the promising approach for WSN power supply, thus, WSN application can be expanded without the need for battery change maintenance or other energy supply systems;
- Novel biosensors can enhance both the number of different sensing molecules and the ability to analyze preliminary data. Then, there are more different parameters we can measure than more useful data for predictive models we can receive [126]. It is significant for surface water and rainwater quality monitoring. Additionally, synthetic biology approaches can help to develop cell-based biosensors which do not use electricity for measurements and processing inside the cell itself;
- Bioactuators can operate partially independently and generate a signal about its state almost because of its biological component activity and without electrical energy consumption.

Thus, biotechnological approaches for sensing and actuation mentioned in this paper give the possibility to increase the amount of data without the energy consumption for sensing and actuation increasing.

Several biotechnological approaches mentioned in this paper are already able to significantly change several aspects of a city in a positive way, producing food and different phototrophic microorganisms' products, treating air and water, and providing a fixation of carbon dioxide. Novel biosensors can significantly improve the capability of WSNs by adding a number of new measuring parameters in combination with MFCs, making them more versatile in terms of energy efficiency.

Thus, it can be concluded that the integration of modern biotechnology approaches in today's cities can provide a significant positive influence on city developments.

Funding: This work was supported by the NRC "Kurchatov Institute", Thematic Plan 1.11, "Development of Nature-Like Bioenergy and Hybrid Energy Sources for Different Application Fields".

Acknowledgments: The author acknowledges colleagues from the Biotechnology and Bioenergy departments for ongoing support.

Conflicts of Interest: The author declares that there is no conflict of interest.

Abbreviations

ANN	Artificial Neural Network
BFC	Biofuel Cells
CPS	Cyber-Physical Systems
HLM	Hybrid Living Materials
IoT	Internet of Things
LPWAN	Low-Power Wide-Area Network
MFC	Microbial Fuel Cell
PMFC	Phototrophic Microbial Fuel Cell
WLAN	Wireless Local Area Network
WSN	Wireless Sensors Networks

References

1. Zhu, C.; Leung, V.C.M.; Shu, L.; Ngai, E.C.H. Green internet of things for smart World. *IEEE Access* **2015**, *3*, 2151–2162. [CrossRef]
2. Zanella, A.; Bui, N.; Castellani, A.; Vangelista, L.; Zorzi, M. Internet of things for smart cities. *IEEE Internet Things J.* **2014**, *1*, 22–32. [CrossRef]
3. Jin, J.; Gubbi, J.; Marusic, S.; Palaniswami, M. An information framework for creating a smart city through internet of things. *IEEE Internet Things J.* **2014**, *1*, 112–121. [CrossRef]
4. Luger, J. Re-envisioning the global city's future. *City* **2019**, *23*, 676–680. [CrossRef]
5. Anzoise, V.; Slanzi, D.; Poli, I. Local stakeholders' narratives about large-scale urban development: The Zhejiang Hangzhou Future Sci-Tech City. *Urban Stud.* **2020**, *57*, 655–671. [CrossRef]
6. Frank, S. Digital natives and living in the City of the future: Contradictions and ambivalences. In *Inequalith and Uncertainty*; Springer: Singapore, 2020; pp. 63–83. [CrossRef]
7. Ragheb, A.; El-Shimy, H.; Ragheb, G. Green Architecture: A Concept of Sustainability. *Procedia Soc. Behav. Sci.* **2016**, *216*, 778–787. [CrossRef]
8. Saha, H.N.; Auddy, S.; Pal, S.; Kumar, S.; Jasu, S.; Singh, R.; Singh, R.; Banerjee, S.; Sharan, P.; Maity, A. Internet of Things (IoT) on bio-technology. In Proceedings of the 2017 8th Annual Industrial Automation and Electromechanical Engineering Conference, Bangkok, Thailand, 16–18 August 2017; pp. 364–369. [CrossRef]
9. Allam, Z. Biotechnology to Render Future Cities as Living and Intelligent Organisms. In *Biotechnology and Future Cities*; Springer: Cham, Switzerland, 2020; pp. 1–15. [CrossRef]
10. Gotovtsev, P.M.; Dyakov, A.V. Biotechnology and Internet of Things for green smart city application. In Proceedings of the 2016 IEEE 3rd World Forum on Internet of Things (WF-IoT), Reston, VA, USA, 12–14 December 2016; pp. 542–546. [CrossRef]
11. Autixier, L.; Mailhot, A.; Bolduc, S.; Madoux-Humery, A.-S.; Galarneau, M.; Prévost, M.; Dorner, S. Evaluating rain gardens as a method to reduce the impact of sewer overflows in sources of drinking water. *Sci. Total Environ.* **2014**, *499*, 238–247. [CrossRef]
12. Church, S.P. Exploring Green Streets and rain gardens as instances of small scale nature and environmental learning tools. *Landsc. Urban Plan.* **2015**, *134*, 229–240. [CrossRef]
13. Al-Chalabi, M. Vertical farming: Skyscraper sustainability? *Sustain. Cities Soc.* **2015**, *18*, 74–77. [CrossRef]
14. Öncel, S.Ş.; Köse, A.; Öncel, D.Ş. 11-Façade integrated photobioreactors for building energy efficiency. In *Start-Up Creat*; Elsevier: Amsterdam, The Netherlands, 2016; pp. 237–299. [CrossRef]
15. Talaei, M.; Mahdavinejad, M. Probable cause of damage to the panel of microalgae bioreactor building façade: Hypothetical evaluation. *Eng. Fail. Anal.* **2019**, *101*, 9–21. [CrossRef]
16. Bhattacharya, M.; Goswami, S. Microalgae—A green multi-product biorefinery for future industrial prospects. *Biocatal. Agric. Biotechnol.* **2020**. [CrossRef]
17. Gorin, K.V.; Sergeeva, Y.E.; Pojidaev, V.M.; Konova, I.A.; Borgolov, A.V.; Gotovtsev, P.M. Thaw water treatment under Moscow insolation conditions by microalgae. *Results Eng.* **2019**, *4*. [CrossRef]
18. Somov, A.; Gotovtsev, P.; Dyakov, A.; Alenicheva, A.; Plehanova, Y.; Tarasov, S.; Reshetilov, A. Bacteria to power the smart sensor applications: Biofuel cell for low-power IoT devices. In Proceedings of the 2018 IEEE 4th World Forum on Internet of Things (WF-IoT), Singapore, 5–8 February 2018; pp. 802–806. [CrossRef]

19. Vishnevskaya, M.; Gazizova, D.; Victorenko, A.; Konova, I. Membraneless microbial biofuel cell for municipal waste water treatment. In *IOP Conference Series: Earth and Environmental Science*; Institute of Physics Publishing: Bristol, UK, 2019. [CrossRef]
20. Jiang, Y.; Yang, X.; Liang, P.; Liu, P.; Huang, X. Microbial fuel cell sensors for water quality early warning systems: Fundamentals, signal resolution, optimization and future challenges, Renew. *Sustain. Energy Rev.* **2018**, *81*, 292–305. [CrossRef]
21. Keating, K.W.; Young, E.M. Synthetic biology for bio-derived structural materials. *Curr. Opin. Chem. Eng.* **2019**, *24*, 107–114. [CrossRef]
22. Gotovtsev, P.M.; Konova, I.A. Synthetic Biology as a Bridge to Integration of Bio Objects into Internet of Things. In Proceedings of the 2019 International Conference on Sensing and Instrumentation in IoT Era (ISSI), Lisbon, Portugal, 29–30 August 2019; pp. 1–6. [CrossRef]
23. Clancy, K.; Voigt, C.A. Programming cells: Towards an automated 'Genetic Compiler'. *Curr. Opin. Biotechnol.* **2010**, *21*, 572–581. [CrossRef] [PubMed]
24. Navarro, P.; Pérez, F.; Weiss, J. Egea-Cortines, M. Machine Learning and Computer Vision System for Phenotype Data Acquisition and Analysis in Plants. *Sensors* **2016**, *16*, 641. [CrossRef]
25. Pölling, B.; Sroka, W.; Mergenthaler, M. Success of urban farming's city-adjustments and business models—Findings from a survey among farmers in Ruhr Metropolis, Germany. *Land Use Policy* **2017**, *69*, 372–385. [CrossRef]
26. Somov, A.; Shadrin, D.; Fastovets, I.; Nikitin, A.; Matveev, S.; Seledets, I.; Hrinchuk, O. Pervasive Agriculture: IoT-Enabled Greenhouse for Plant Growth Control. *IEEE Pervasive Comput.* **2018**, *17*, 65–75. [CrossRef]
27. Song, X.P.; Tan, H.T.W.; Tan, P.Y. Assessment of light adequacy for vertical farming in a tropical city. *Urban. For. Urban Green.* **2018**, *29*, 49–57. [CrossRef]
28. Pappu, J.S.M.; Vijayakumar, G.K.; Ramamurthy, V. Artificial neural network model for predicting production of Spirulina platensis in outdoor culture. *Bioresour. Technol.* **2013**, *130*, 224–230. [CrossRef]
29. Supriyanto, R.; Noguchi, T.; Ahamed, D.S.; Rani, K.; Sakurai, M.A.; Nasution, D.S.; Wibawa, M.; Demura, M.; Watanabe, M. Artificial neural networks model for estimating growth of polyculture microalgae in an open raceway pond. *Biosyst. Eng.* **2019**, *177*, 122–129. [CrossRef]
30. Logan, B.E.; Regan, J.M. Electricity-producing bacterial communities in microbial fuel cells. *Trends Microbiol.* **2006**, *14*, 512–518. [CrossRef]
31. Naraghi, Z.G.; Yaghmaei, S.; Mardanpour, M.M.; Hasany, M. Produced Water Treatment with Simultaneous Bioenergy Production Using Novel Bioelectrochemical Systems. *Electrochim. Acta* **2015**, *180*, 535–544. [CrossRef]
32. Gotovtsev, P.; Vorobiev, V.; Migalev, A.; Badranova, G.; Gorin, K.; Dyakov, A.; Reshetilov, A. Bioenergy Based Power Sources for Mobile Autonomous Robots. *Robotics* **2018**, *7*, 2. [CrossRef]
33. Santoro, C.; Arbizzani, C.; Erable, B.; Ieropoulos, I. Microbial fuel cells: From fundamentals to applications. A review. *J. Power Sources* **2017**, *356*, 225–244. [CrossRef]
34. Deng, H.; Chen, Z.; Zhao, F. Energy from Plants and Microorganisms: Progress in Plant-Microbial Fuel Cells. *ChemSusChem* **2012**, *5*, 1006–1011. [CrossRef]
35. Greenman, J.; Gajda, I.; Ieropoulos, I. Microbial fuel cells (MFC) and microalgae; photo microbial fuel cell (PMFC) as complete recycling machines. *Sustain. Energy Fuels* **2019**, *3*, 2546–2560. [CrossRef]
36. Helder, M.; Strik, D.P.B.T.B.; Hamelers, H.V.M.; Kuhn, A.J.; Blok, C.; Buisman, C.J.N. Concurrent bio-electricity and biomass production in three Plant-Microbial Fuel Cells using Spartina anglica, Arundinella anomala and Arundo donax. *Bioresour. Technol.* **2010**, *101*, 3541–3547. [CrossRef]
37. Pant, D.; van Bogaert, G.; Diels, L.; Vanbroekhoven, K. A review of the substrates used in microbial fuel cells (MFCs) for sustainable energy production. *Bioresour. Technol.* **2010**, *101*, 1533–1543. [CrossRef]
38. Reshetilov, A.N.; Plekhanova, Y.V.; Tarasov, S.E.; Arlyapov, V.A.; Kolesov, V.V.; Gutorov, M.A.; Gotovtsev, P.M.; Vasilov, R.G. Effect of some carbon nanomaterials on ethanol oxidation by Gluconobacter oxydans bacterial cells. *Appl. Biochem. Microbiol.* **2017**, *53*, 123–129. [CrossRef]
39. Reshetilov, A.N.; Kitova, A.E.; Dyakov, A.V.; Gotovtsev, P.M.; Vasilov, R.G.; Gutorov, M.A. Converter-based accumulation of electric energy generated by microbial biofuel cell. *IOP Conf. Ser. Earth Environ. Sci.* **2017**, *52*. [CrossRef]
40. Choi, S.; Lee, H.-S.; Yang, Y.; Parameswaran, P.; Torres, C.I.; Rittmann, B.E.; Chae, J. A µL-scale micromachined microbial fuel cell having high power density. *Lab Chip.* **2011**, *11*, 1110–1117. [CrossRef] [PubMed]

41. Kaneshiro, H.; Takano, K.; Takada, Y.; Wakisaka, T.; Tachibana, T.; Azuma, M. A milliliter-scale yeast-based fuel cell with high performance. *Biochem. Eng. J.* **2014**, *83*, 90–96. [CrossRef]
42. Liu, X.; Wu, W.; Gu, Z. Poly (3,4-ethylenedioxythiophene) promotes direct electron transfer at the interface between Shewanella loihica and the anode in a microbial fuel cell. *J. Power Sources* **2015**, *277*, 110–115. [CrossRef]
43. Huarachi-Olivera, R.; Dueñas-Gonza, A.; Yapo-Pari, U.; Vega, P.; Romero-Ugarte, M.; Tapia, J.; Molina, L.; Lazarte-Rivera, A.; Pacheco-Salazar, D.G.; Esparza, M. Bioelectrogenesis with microbial fuel cells (MFCs) using the microalga Chlorella vulgaris and bacterial communities. *Electron. J. Biotechnol.* **2018**, *31*, 34–43. [CrossRef]
44. Hashemi, N.; Lackore, J.M.; Sharifi, F.; Goodrich, P.J.; Winchell, M.L.; Hashemi, N. A paper-based microbial fuel cell operating under continuous flow condition. *Technology* **2016**, *04*, 98–103. [CrossRef]
45. Ren, H.; Torres, C.I.; Parameswaran, P.; Rittmann, B.E.; Chae, J. Improved current and power density with a micro-scale microbial fuel cell due to a small characteristic length. *Biosens. Bioelectron.* **2014**, *61*, 587–592. [CrossRef]
46. Angioni, E.Q.S.; Millia, L.; Mustarelli, P.; Doria, E.; Temporiti, M.E.; Mannucci, B.; Corana, F. Photosynthetic microbial fuel cell with polybenzimidazole membrane: Synergy between bacteria and algae for wastewater removal and biorefinery. *Heliyon* **2018**, *4*. [CrossRef]
47. Mohamed, S.N.; Hiraman, P.A.; Muthukumar, K.; Jayabalan, T. Bioelectricity production from kitchen wastewater using microbial fuel cell with photosynthetic algal cathode. *Bioresour. Technol.* **2020**, *295*. [CrossRef]
48. Yang, N.; Zhan, G.; Li, D.; Wang, X.; He, X.; Liu, H. Complete nitrogen removal and electricity production in Thauera-dominated air-cathode single chambered microbial fuel cell. *Chem. Eng. J.* **2019**, *356*, 506–515. [CrossRef]
49. Zhang, M.; Ma, Z.; Zhao, N.; Zhang, K.; Song, H. Increased power generation from cylindrical microbial fuel cell inoculated with P. aeruginosa. *Biosens. Bioelectron.* **2019**, *141*. [CrossRef] [PubMed]
50. Nguyen, V.; Nitisoravut, R. Bioelectricity Generation in Plant Microbial Fuel Cell Using Forage Grass under Variations of Circadian Rhythm, Ambient Temperature, and Soil Water Contents. In Proceedings of the 2019 IEEE Asia Power and Energy Engineering Conference (APEEC), Chengdu, China, 29–31 March 2019; pp. 240–244. [CrossRef]
51. de la Rosa, E.O.; Castillo, J.V.; Campos, M.C.; Pool, G.B.; Nuñez, G.B.; Atoche, A.C.; Aguilar, J.O. Plant Microbial Fuel Cells–Based Energy Harvester System for Self-powered IoT Applications. *Sensors* **2019**, *19*, 1378. [CrossRef] [PubMed]
52. Reshetilov, A.N. Biosensors and biofuel cells: Research focused on practical application (Review). *Appl. Biochem. Microbiol.* **2015**, *51*, 264–269. [CrossRef]
53. Oliot, M.; Etcheverry, L.; Mosdale, R.; Bergel, A. Microbial fuel cells connected in series in a common electrolyte underperform: Understanding why and in what context such a set-up can be applied. *Electrochim. Acta* **2017**, *246*, 879–889. [CrossRef]
54. Ikpehai, A.; Adebisi, B.; Rabie, K.M.; Anoh, K.; Ande, R.E.; Hammoudeh, M.; Gacanin, H.; Mbanaso, U.M. Low-power wide area network technologies for internet-of-things: A comparative review. *IEEE Internet Things J.* **2019**, *6*, 2225–2240. [CrossRef]
55. Qadir, Q.M.; Rashid, T.A.; Al-Salihi, N.K.; Ismael, B.; Kist, A.A.; Zhang, Z. Low power wide area networks: A survey of enabling technologies, applications and interoperability needs. *IEEE Access* **2018**, *6*, 77454–77473. [CrossRef]
56. Salwe, S.S.; Naik, K.K. HeterogeneousWireless Network for IoT Applications. *IETE Tech. Rev. (Inst. Electron. Telecommun. Eng. India)* **2019**, *36*, 61–68. [CrossRef]
57. Monteiro, A.; Souto, E.; Pazzi, R.; Nogueira, M. Context-aware network selection in heterogeneous wireless networks. *Comput. Commun* **2019**, *135*, 1–15. [CrossRef]
58. Do, M.H.; Ngo, H.H.; Guo, W.; Chang, S.W.; Nguyen, D.D.; Liu, Y.; Varjani, S.; Kumar, M. Microbial fuel cell-based biosensor for online monitoring wastewater quality: A critical review. *Sci. Total Environ.* **2020**, *172*, 135612. [CrossRef]
59. Grattieri, M.; Minteer, S.D. Self-Powered Biosensors. *ACS Sens.* **2018**, *3*, 44–53. [CrossRef]

60. Sarkar, A.; Sarkar, K.D.; Amrutha, V.; Dutta, K. An overview of enzyme-based biosensors for environmental monitoring. In *Tools, Techniques and Protocols for Monitoring Environmental Contaminants*; Elsevier: Amsterdam, The Netherlands, 2019; pp. 307–329. [CrossRef]
61. Gupta, N.; Renugopalakrishnan, V.; Liepmann, D.; Paulmurugan, R.; Malhotra, B.D. Cell-based biosensors: Recent trends, challenges and future perspectives. *Biosens. Bioelectron.* **2019**, *141*. [CrossRef]
62. Taketani, M.; Zhang, J.; Zhang, S.; Triassi, A.J.; Huang, Y.-J.; Griffith, L.G.; Voigt, C.A. Genetic circuit design automation for the gut resident species Bacteroides thetaiotaomicron. *Nat. Biotechnol.* **2020**, 1–8. [CrossRef]
63. Vaidyanathan, P.; Der, B.S.; Bhatia, S.; Roehner, N.; Silva, R.; Voigt, C.a.; Densmore, D. A Framework for Genetic Logic Synthesis. *Proc. IEEE* **2015**, *103*, 2196–2207. [CrossRef]
64. Adekunle, A.; Raghavan, V.; Tartakovsky, B. On-line monitoring of heavy metals-related toxicity with a microbial fuel cell biosensor. *Biosens. Bioelectron.* **2019**, *132*, 382–390. [CrossRef]
65. Bilal, M.; Iqbal, H.M.N. Microbial-derived biosensors for monitoring environmental contaminants: Recent advances and future outlook. *Process Saf. Environ. Prot.* **2019**, *124*, 8–17. [CrossRef]
66. Jiang, Y.; Liang, P.; Liu, P.; Wang, D.; Miao, B.; Huang, X. A novel microbial fuel cell sensor with biocathode sensing element. *Biosens. Bioelectron.* **2017**, *94*, 344–350. [CrossRef]
67. Halilovic, A.; Merdan, E.; Kovacevic, Z.; Pokvic, L.G. Review of Biosensors for Environmental Field Monitoring. In Proceedings of the 2019 8th Mediterranean Conference on Embedded Computing (MECO), Budva, Montenegro, 10–14 June 2019. [CrossRef]
68. Damborský, P.; Švitel, J.; Katrlík, J.; Dey, D.; Goswami, T.; Liedberg, B.; Nylander, C.; Lunström, I.; Schasfoort, R.B.M.; Tudos, A.J.; et al. Optical biosensors. *Essays Biochem.* **2016**, *60*, 91–100. [CrossRef]
69. Nielsen, A.K.; Der, B.S.; Shin, J.; Vaidyanathan, P.; Densmore, D.; Voigt, C.a. Genetic circuit design automation. *Science (80-.)* **2016**, *352*, 53–64. [CrossRef]
70. Zhou, X.; Franklin, R.A.; Adler, M.; Jacox, J.B.; Bailis, W.; Shyer, J.A.; Flavell, R.A.; Mayo, A.; Alon, U.; Medzhitov, R. Circuit Design Features of a Stable Two-Cell System. *Cell* **2018**, *172*, 744–757. [CrossRef]
71. Fernandez-Rodriguez, J.; Moser, F.; Song, M.; Voigt, C.A. Engineering RGB color vision into Escherichia coli. *Nat. Chem. Biol.* **2017**, *13*, 706–708. [CrossRef]
72. Shin, J.; Zhang, S.; Der, B.S.; Nielsen, A.A.; Voigt, C.A. Programming *Escherichia coli* to function as a digital display. *Mol. Syst. Biol.* **2020**, *16*, 1–12. [CrossRef]
73. Grunberg, T.W.; del Vecchio, D. Modular Analysis and Design of Biological Circuits. *Curr. Opin. Biotechnol.* **2020**, *63*, 41–47. [CrossRef]
74. Torrance, A.W. Synthesizing Law for Synthetic Biology. *Minnesota J. Law Sci. Technol.* **2010**, *11*. Available online: https://heinonline.org/HOL/Page?handle=hein.journals/mipr11&id=629&div=&collection= (accessed on 9 April 2020).
75. Lauffenburger, D.A. Cell signaling pathways as control modules: Complexity for simplicity? *Proc. Natl. Acad. Sci. USA* **2000**, *97*, 5031–5033. [CrossRef]
76. del Vecchio, D.; Ninfa, A.J.; Sontag, E.D. Modular cell biology: Retroactivity and insulation. *Mol. Syst. Biol.* **2008**, *4*, 161. [CrossRef]
77. del Vecchio, D.; Abdallah, H.; Qian, Y.; Collins, J.J. A Blueprint for a Synthetic Genetic Feedback Controller to Reprogram Cell Fate. *Cell Syst.* **2017**, *4*, 109–120. [CrossRef]
78. Sarpeshkar, R. Analog synthetic biology. *Philos. Trans. Math. Phys. Eng. Sci.* **2014**, *372*. [CrossRef]
79. Banerjee, A.; Weaver, I.; Thorsen, T.; Sarpeshkar, R. Bioelectronic measurement and feedback control of molecules in living cells. *Sci. Rep.* **2017**, *7*. [CrossRef]
80. Zeng, J.; Kim, J.; Banerjee, A.; Sarpeshkar, R. A Synthetic Microbial Operational Amplifier. *ACS Synth. Biol.* **2018**. [CrossRef]
81. Cotton, G.J.; Ayers, B.; Xu, R.; Muir, T.W. Insertion of a synthetic peptide into a recombinant protein framework: A protein biosensor. *J. Am. Chem. Soc.* **1999**, *121*, 1100–1101. [CrossRef]
82. Yu, K.; Liu, C.; Kim, B.G.; Lee, D.Y. Synthetic fusion protein design and applications. *Biotechnol. Adv.* **2015**, *33*, 155–164. [CrossRef]
83. Lahoz-Beltra, R.; Hameroff, S.R.; Dayhoff, J.E. Cytoskeletal logic: A model for molecular computation via Boolean operations in microtubules and microtubule-associated proteins. *BioSystems* **1993**, *29*, 1–23. [CrossRef]
84. Katz, E. Boolean Logic Gates Realized with Enzyme-catalyzed Reactions-Unusual Look at Usual Chemical Reactions. *Chem. Phys. Chem.* **2019**, *20*, 9–22. [CrossRef]

85. Chen, Z.; Kibler, R.D.; Hunt, A.; Busch, F.; Pearl, J.; Jia, M.; Vanaernum, Z.L.; Wicky, B.I.M.; Dods, G.; Liao, H.; et al. De novo design of protein logic gates. *Science (80-.)* **2020**, *84*, 78–84. [CrossRef]
86. Bianchini, F. Artificial intelligence and synthetic biology: A tri-temporal contribution. *Biosystems* **2016**. [CrossRef]
87. Shih, C.-S.; Chou, J.-J.; Reijers, N.; Kuo, T.-W. Designing CPS/IoT applications for smart buildings and cities. *IET Cyber-Phys. Syst. Theory Appl.* **2016**, *1*, 3–12. [CrossRef]
88. Karr, J.R.; Sanghvi, J.C.; MacKlin, D.N.; Gutschow, M.V.; Jacobs, J.M.; Bolival, B.; Assad-Garcia, N.; Glass, J.I.; Covert, M.W. A whole-cell computational model predicts phenotype from genotype. *Cell* **2012**, *150*, 389–401. [CrossRef]
89. Daniel, R.; Rubens, J.R.; Sarpeshkar, R.; Lu, T.K. Synthetic analog computation in living cells. *Nature* **2013**, *497*, 619–623. [CrossRef]
90. Teo, J.J.Y.; Woo, S.S.; Sarpeshkar, R. Synthetic Biology: A Unifying View and Review Using Analog Circuits. *IEEE Trans. Biomed. Circuits Syst.* **2015**, *9*, 453–474. [CrossRef]
91. Kobusińska, A.; Leung, C.; Hsu, C.-H.; Chang, S.R.V. Emerging trends, issues and challenges in Internet of Things, Big Data and cloud computing. *Future Gener. Comput. Syst.* **2018**, *87*, 416–419. [CrossRef]
92. Stankovic, J.A. Research Directions for the Internet of Things. *IEEE Internet Things J.* **2014**, *1*, 3–9. [CrossRef]
93. Yoon, C.; Huh, M.; Kang, S.G.; Park, J.; Lee, C. Implement smart farm with IoT technology. In Proceedings of the 2018 20th International Conference on Advanced Communication Technology (ICACT), Gangwon-do, Korea, 11–14 February 2018; pp. 749–752. [CrossRef]
94. Zhang, Y.; Chung, T.S. Graphene oxide membranes for nanofiltration. *Curr. Opin. Chem. Eng.* **2017**, *16*, 9–15. [CrossRef]
95. Liu, X.; Yuk, H.; Lin, S.; Parada, G.A.; Tang, T.C.; Tham, E.; de la Fuente-Nunez, C.; Lu, T.K.; Zhao, X. 3D Printing of Living Responsive Materials and Devices. *Adv. Mater.* **2018**, *30*. [CrossRef] [PubMed]
96. Liu, X.; Tang, T.C.; Tham, E.; Yuk, H.; Lin, S.; Lu, T.K.; Zhao, X. Stretchable living materials and devices with hydrogel-elastomer hybrids hosting programmed cells. *Proc. Natl. Acad. Sci. USA* **2017**, *114*, 2200–2205. [CrossRef]
97. le Feuvre, R.A.; Scrutton, N.S. A living foundry for Synthetic Biological Materials: A synthetic biology roadmap to new advanced materials. *Synth. Syst. Biotechnol.* **2018**, *3*, 105–112. [CrossRef] [PubMed]
98. Smith, R.S.H.; Bader, C.; Sharma, S.; Kolb, D.; Tang, T.C.; Hosny, A.; Moser, F.; Weaver, J.C.; Voigt, C.A.; Oxman, N. Hybrid Living Materials: Digital Design and Fabrication of 3D Multimaterial Structures with Programmable Biohybrid Surfaces. *Adv. Funct. Mater.* **2020**, *30*. [CrossRef]
99. Wagner, H.J.; Engesser, R.; Ermes, K.; Geraths, C.; Timmer, J.; Weber, W. Synthetic biology-inspired design of signal-amplifying materials systems. *Mater. Today* **2019**, *22*, 25–34. [CrossRef]
100. Chen, X.; Goodnight, D.; Gao, Z.; Cavusoglu, A.H.; Sabharwal, N.; DeLay, M.; Driks, A.; Sahin, O. Scaling up nanoscale water-driven energy conversion into evaporation-driven engines and generators. *Nat. Commun.* **2015**, *6*. [CrossRef] [PubMed]
101. Shadrin, D.; Menshchikov, A.; Ermilov, D.; Somov, A. Designing Future Precision Agriculture: Detection of Seeds Germination Using Artificial Intelligence on a Low-Power Embedded System. *IEEE Sens. J.* **2019**, *19*, 11573–11582. [CrossRef]
102. Shadrin, D.; Menshchikov, A.; Somov, A.; Bornemann, G.; Hauslage, J.; Fedorov, M. Enabling Precision Agriculture through Embedded Sensing with Artificial Intelligence. *IEEE Trans. Instrum. Meas.* **2019**, *9456*, 1. [CrossRef]
103. Erlich, Y.; Zielinski, D. DNA Fountain enables a robust and efficient storage architecture. *Science (80-.)* **2017**, *355*, 950–954. [CrossRef] [PubMed]
104. Church, G.M.; Gao, Y.; Kosuri, S. Next-generation digital information storage in DNA. *Science (80-.)* **2012**, *337*, 1628. [CrossRef] [PubMed]
105. Koch, J.; Gantenbein, S.; Masania, K.; Stark, W.J.; Erlich, Y.; Grass, R.N. A DNA-of-things storage architecture to create materials with embedded memory. *Nat. Biotechnol.* **2020**, *38*, 39–43. [CrossRef] [PubMed]
106. Aitken, M.; Toreini, E.; Carmichael, P.; Coopamootoo, K.; Elliott, K.; van Moorsel, A. Establishing a social licence for Financial Technology: Reflections on the role of the private sector in pursuing ethical data practices. *Big Data Soc.* **2020**, *7*. [CrossRef]

107. Boles, K.S.; Kannan, K.; Gill, J.; Felderman, M.; Gouvis, H.; Hubby, B.; Kamrud, K.I.; Venter, J.C.; Gibson, D.G. Digital-to-biological converter for on-demand production of biologics. *Nat. Biotechnol.* **2017**, *35*, 672–675. [CrossRef] [PubMed]
108. Norville, J.E.; Derda, R.; Gupta, S.; Drinkwater, K.A.; Belcher, A.M.; Leschziner, A.E.; Knight, T.F. Introduction of customized inserts for streamlined assembly and optimization of BioBrick synthetic genetic circuits. *J. Biol. Eng.* **2010**, *4*, 17. [CrossRef]
109. Galdzicki, M.; Rodriguez, C.; Chandran, D.; Sauro, H.M.; Gennari, J.H. Standard Biological Parts Knowledgebase. *PLoS ONE.* **2011**, *6*. [CrossRef]
110. Connell, J.L.; Ritschdorff, E.T.; Whiteley, M.; Shear, J.B. 3D printing of microscopic bacterial communities. *Proc. Natl. Acad. Sci. USA* **2013**, *110*, 18380–18385. [CrossRef]
111. Hsu, L.; Jiang, X. 'Living' Inks for 3D Bioprinting. *Trends Biotechnol.* **2019**. [CrossRef]
112. Libbrecht, M.W.; Noble, W.S. Machine learning applications in genetics and genomics. *Nat. Rev. Genet.* **2015**, *16*, 321–332. [CrossRef]
113. Nielsen, A.A.K.; Voigt, C.A. Deep learning to predict the lab-of-origin of engineered DNA. *Nat. Commun.* **2018**, *9*, 3135. [CrossRef]
114. Cheng, J.; Tegge, A.N.; Baldi, P. Machine Learning Methods for Protein Structure Prediction. *IEEE Rev. Biomed. Eng.* **2008**, *1*, 41–49. [CrossRef]
115. Costello, Z.; Martin, H.G. A machine learning approach to predict metabolic pathway dynamics from time-series multiomics data. *NPJ Syst. Biol. Appl.* **2018**, *4*, 19. [CrossRef]
116. Shadrin, D.; Somov, A.; Podladchikova, T.; Gerzer, R. Pervasive agriculture: Measuring and predicting plant growth using statistics and 2D/3D imaging. In Proceedings of the 2018 IEEE International Instrumentation and Measurement Technology Conference (I2MTC), Houston, TX, USA, 14–17 May 2018; pp. 1–6. [CrossRef]
117. Hu, D.; Liu, H.; Yang, C.; Hu, E. The design and optimization for light-algae bioreactor controller based on Artificial Neural Network-Model Predictive Control. *Acta Astronaut.* **2008**, *63*, 1067–1075. [CrossRef]
118. García-Camacho, F.; López-Rosales, L.; Sánchez-Mirón, A.; Belarbi, E.H.; Chisti, Y.; Molina-Grima, E. Artificial neural network modeling for predicting the growth of the microalga Karlodinium veneficum. *Algal Res.* **2016**, *14*, 58–64. [CrossRef]
119. del Rio-Chanona, E.A.; Manirafasha, E.; Zhang, D.; Yue, Q.; Jing, K. Dynamic modeling and optimization of cyanobacterial C-phycocyanin production process by artificial neural network. *Algal Res.* **2016**, *13*, 7–15. [CrossRef]
120. Pruvost, J.; le Gouic, B.; Lepine, O.; Legrand, J.; le Borgne, F. Microalgae culture in building-integrated photobioreactors: Biomass production modelling and energetic analysis. *Chem. Eng. J.* **2015**, *284*, 850–861. [CrossRef]
121. Yuan, Y.; Yu, X.; Yang, X.; Xiao, Y.; Xiang, B.; Wang, Y. Bionic building energy efficiency and bionic green architecture: A review. *Renew. Sustain. Energy Rev.* **2017**, *74*, 771–787. [CrossRef]
122. Petrolo, R.; Loscri, V.; Mitton, N. Cyber-physical objects as key elements for a smart cyber-city. In *Internet of Things*; Springer: Berlin, Germany, 2016; pp. 31–49. [CrossRef]
123. Buhk, H.J. Synthetic biology and its regulation in the European Union. *New Biotechnol.* **2014**, *31*, 528–531. [CrossRef]
124. Ahteensuu, M. Synthetic Biology, Genome Editing, and the Risk of Bioterrorism. *Sci. Eng. Ethics* **2017**, *23*, 1541–1561. [CrossRef]
125. Oltvai, Z.N.; Barabási, A.-L. Systems biology. Life's complexity pyramid. *Science* **2002**, *298*, 763–764. [CrossRef] [PubMed]
126. Zhu, C.; Zhou, H.; Leung, V.C.M.; Wang, K.; Zhang, Y.; Yang, L.T. Toward Big Data in Green City. *IEEE Commun. Mag.* **2017**, *55*, 14–18. [CrossRef]