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Biosorption of Lithium using Microbes

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BIOSORPTION OF LITHIUM USING MICROBES

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Lithium (Li) is a very valuable metal that is used across several industries including ceramics, glass, batteries, pharmaceuticals, and polymers. However, in recent years, the global demand for Li and its market price have increased considerably, due to its application as a critical component in the production of rechargeable Li-ion batteries and energy storage systems that are used in electric vehicles and a variety of electronic devices. Although Li occurs as a mineral in hard rocks and salt brines, substantial amounts are found in our environment as part of industrial wastes and oil-field wastewaters. Despite its importance, Li is also harmful and poses a risk to the environment. Besides, the conventional (chemical and physical) methods that are used today for its removal, such as solvent extraction and acid leaching, require high energy consumption and produce toxic by-products, posing additional environmental and economic challenges. Alternatively, the use of bacteria for Li extraction has been proposed as a viable, non-toxic, and cost-effective alternative. In this study, the potential of using Gram-negative *Escherichia coli*, and Gram-positive *Bacillus subtilis* and *Bacillus cereus* as biosorbents for Li was explored. Results indicate that all three bacterial species tested were capable of absorbing Li to

varying degrees from aqueous solutions. However, *E. coli* had the highest and most consistent absorption capacity and was selected for further investigation. The amounts of total dissolved solids (TDS) and Li, analyzed by inductively coupled plasma atomic emission spectroscopy (ICP-OES), methods were obtained in this study. In a kinetic study of Li biosorption, most Li-binding occurred within the first 24h and slowed down until maximum biosorption was attained following 72h, our experimental endpoint. The biosorption capacity for *E. coli* ranged from 60% to 43% depending on initial Li concentrations in solution. Also, the optimal pH for E. coli biosorption was found to be between pH 6-6.5. Recovered/eluted absorbed Li was measured following a 12h mild-acid solution (distilled H_2O adjusted to pH 4 with HNO_3) wash of the Li-bound biomass membranes.

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ICP-OES	Inductively Coupled Plasma Optical Emission Spectroscopy
pH	Hydrogen ion Concentration
PPM	Parts Per Million
TDS	Total Dissolved Solids

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CHAPTER 1

INTRODUCTION

Biosorption is the ability of biological materials to bind and accumulate heavy metals from aqueous solutions such as wastewater. It is a remarkable and intricate process that demonstrates the extraordinary ability of living organisms to assimilate and merge substances from their environment into their very own biological structures. This extraordinary phenomenon spans across various levels of biology, ranging from the cellular level to the tissue and organism levels. At the cellular level, biosorption involves the absorption of nutrients, gases, and other molecules by cells to support their metabolic functions and promote growth. Tissues, on the other hand, undergo biosorption as they assimilate substances for the purpose of repair, regeneration, and the maintenance of their structural integrity (Tsuruta, 2005). On a much grander scale, organisms engage in the interesting process of biosorption in order to extract essential elements or compounds from their surroundings. This process, is undeniably fundamental to the sustenance and development of living entities, serves as a catalyst for the conversion of external resources into internal building blocks.

It is important to note that biosorption is not solely confined to nutritional aspects; it extends its activities to the assimilation of pharmaceuticals or biomaterials in the field of medicine, where the body skillfully absorbs and incorporates foreign substances for therapeutic purposes (Yun and Volesky, 2003). Appreciating the intricacies of bio absorption is pivotal in various scientific disciplines, including biology, medicine, and environmental science, as it enlightens us about the dynamic interplay between living organisms and their ever-changing surroundings (Sethurajan and Gaydardzhiev, 2021).

Lithium plays a pivotal role in rechargeable lithium-ion batteries, driving the portable electronics revolution. These batteries are crucial for powering devices like smartphones, laptops, and electric vehicles, owing to their high energy density and long-lasting performance. In the pharmaceutical industry, lithium compounds are utilized for their mood-stabilizing properties. Lithium is a key component in medications prescribed to individuals with bipolar disorder, aiding in the regulation of neurotransmitter activity in the brain (Dolker & Pant, 2019).

Lithium's lightweight characteristics make it an essential element in the aerospace sector. It is employed in the production of lightweight alloys, contributing to the construction of aircraft and spacecraft components. These alloys enhance fuel efficiency and overall performance. Lithium grease, derived from lithium soap, serves as a widely used lubricating material. Its resistance to water and high temperatures makes it indispensable in automotive and industrial applications, ensuring smooth machinery operation (Huang, Liu, and Zhang, 2019). In nuclear reactors, lithium acts as a neutron moderator, controlling nuclear reactions. Its role in nuclear technology is critical for ensuring safe and controlled energy production. Lithium compounds find applications in various industrial processes, such as ceramics and glass production, as well as serving as a flux in metal processing. The versatility of lithium contributes significantly to modern manufacturing and material development (Heydarian et al., 2018).

Lithium possesses a set of extraordinary physical and chemical properties that distinguish it from other metals. As the lightest alkali metal, it has the lowest atomic number and density among solid elements. This exceptional metal plays an indispensable role in rechargeable batteries, particularly lithium-ion batteries, which power a vast range of electronic gadgets and electric vehicles. Its remarkable electrochemical characteristics, stemming from its impressive electrochemical potential and low atomic weight, greatly enhance its efficacy in storing and discharging electrical energy (Horeb et al., 2016). Moreover, lithium shows an unparalleled reactivity when it encounters water, leading to the formation of lithium hydroxide and hydrogen gas. This unique trait sets it apart from its alkali metal counterparts, as it exhibits a milder reactivity, making it safer for specific applications. The distinctive behavior of lithium in ion exchange processes, as well as its ability to intercalate within materials, grants it undeniable value in a myriad of fields, including medicine and nuclear physics. Furthermore, lithium possesses the captivating attribute of being one of the rare elements produced in significant quantities through cosmic nucleosynthesis. Its celestial origins further elevate its significance, distinguishing it both on Earth and in the cosmos (Horeh et al., 2016).

The conventional means of extracting lithium typically involves the mining and processing of lithium-containing ores. Lithium is predominantly extracted from two main types of deposits: lithium-rich brine deposits and lithium-containing mineral deposits. Brine deposits are found in salt flats or lakes, primarily in countries like Chile, Argentina, and Bolivia. The process involves pumping lithium-rich brine to the surface from underground reservoirs. The brine is then left to evaporate in large ponds, concentrating the lithium content. Once concentrated, lithium is extracted through a series of chemical processes, including precipitation and ion exchange, resulting in lithium carbonate or lithium hydroxide (Yang et al., 2016). Spodumene and lepidolite are two common lithium-containing minerals that are mined for lithium extraction.

The minerals are typically crushed and subjected to various physical and chemical processes to extract lithium. After mining, the ore undergoes beneficiation processes to concentrate the lithium content. Subsequent roasting and acid treatment are employed to extract lithium in the form of lithium carbonate or lithium hydroxide. These conventional extraction methods are well-established but may involve complex processes, and the environmental impact of mining and processing is a subject of concern. Efforts are ongoing to explore more sustainable and environmentally friendly methods for lithium extraction as the demand for lithium continues to rise in various industries, especially in the production of batteries for electric vehicles and renewable energy storage (Xin et al., 2009).

The exploration of alternative approaches for extracting lithium arises from a multitude of factors, encompassing both environmental considerations and the escalating demand for lithium-driven technologies. Allow me to present to you the key drivers behind the search for alternative methods of lithium extraction. Conventional processes for extracting lithium, particularly from brine deposits, often necessitate the creation of extensive evaporation ponds, which can have profound environmental ramifications such as excessive water consumption, disruption of natural habitats, and the potential for chemical contamination. The extraction of lithium-containing minerals through mining can result in the devastation of forests, destruction of habitats, and the release of hazardous byproducts (Bahaloo et al., 2018). Traditional extraction methods can be excessively energy-intensive, particularly during the heating and chemical processing stages required to obtain lithium carbonate or lithium hydroxide. The environmental consequences of energy consumption, particularly if derived from non-renewable sources, contribute to the overall carbon footprint of lithium production.

The demand for lithium is experiencing an unprecedented surge, driven by the soaring popularity of electric vehicles, renewable energy storage, and portable electronic devices. Consequently, there is a pressing need to diversify lithium sources in order to diminish reliance on a select few countries that dominate its production.

The global shift towards a more sustainable and circular economy has sparked a profound interest in recycling lithium-ion batteries and exploring methods to recover lithium from spent batteries. This strategic move not only reduces dependence on new mining endeavors but also alleviates concerns regarding resource depletion (Vendruscolo et al., 2017). Unceasing research and technological advancements in materials science and chemistry have given rise to the exploration of innovative extraction methods that hold the promise of being more efficient, cost-effective, and environmentally benign.

Traditional lithium extraction activities can significantly impact local communities, raising concerns about water usage, disruption of land, and potential adverse health effects. The pursuit of alternative methods that minimize such impacts is imperative to ensure responsible and ethical practices in lithium production. In summary, the quest for alternative methods of lithium extraction is driven by an unwavering commitment to mitigate the environmental impact, curtail energy consumption, fortify supply chain security, foster sustainability, and address social and community concerns. The development of extraction processes that are both eco-friendly and highly efficient is paramount to ensuring the long-term viability of lithium-based technologies (González-Gil 2011).

The microbial extraction of lithium has garnered considerable attention as a potential alternative to conventional methods, thanks to its impressive environmental sustainability, energy efficiency, and capacity for minimizing ecological impact. Unlike the energy-intensive and environmentally harmful processes involved in conventional means of extraction, such as brine evaporation and mineral processing, microbial extraction embraces the metabolic activities of specific microorganisms to selectively leach lithium from ores or brines. This approach offers a more environmentally friendly and economically viable solution. One of the remarkable advantages of microbial processes is their ability to operate under mild conditions, significantly reducing the reliance on high-temperature roasting or extensive chemical treatments. This not only conserves energy but also minimizes the release of greenhouse gas emissions, which are typically associated with conventional extraction methods (Jin et al., 2017). Additionally, microbial extraction possesses inherent selectivity, enabling the targeted recovery of lithium without co-extracting unwanted impurities.

This streamlines downstream processing efficiently. In certain cases, specific bacteria or fungi can solubilize lithium from minerals through their metabolic activity, making it more easily accessible for subsequent recovery. These microorganisms, typically adapted to extreme environments, release organic acids or other compounds that facilitate the breakdown of the mineral structure and the liberation of lithium ions. Furthermore, in the realm of recycling, microbes can also contribute to the extraction of lithium from spent batteries. This approach aligns perfectly with the principles of a circular economy, as it allows for the recovery of valuable resources from waste materials, ultimately reducing the demand for new mining activities (Torres, 2020).

Microbial extraction of lithium presents a highly promising avenue for sustainable resource recovery, perfectly aligning with global efforts to transition towards greener technologies. However, ongoing research and development are crucial to optimizing microbial processes, improving efficiency, and scaling up operations for industrial applications. As technology continues to advance and our understanding of microbial interactions deepens, microbial extraction may increasingly emerge as a viable and environmentally friendly method of obtaining lithium, thereby addressing the environmental concerns associated with conventional extraction methods (Wang et al., 2019).

Microbes utilized in the extraction of metals possess exceptional qualities that render them highly suitable for this important task. One vital attribute is their extraordinary metabolic versatility, which allows them to effortlessly adapt to a wide array of environmental conditions. These remarkable microorganisms are often extremophiles, thriving in environments with extreme temperatures, highly acidic or alkaline pH levels, or even in areas with high salinity. This remarkable adaptability empowers them to function in diverse settings, ranging from acidic mine environments to the depths of deep-sea hydrothermal vents.

Furthermore, microbes employed in metal extraction typically exhibit either chemoorganotrophic or chemo lithotrophic metabolic pathways. Chemoorganotrophic microbes derive their energy from organic compounds, while chemolithotroph ones obtain their energy from inorganic sources. This exceptional metabolic flexibility allows them to utilize a variety of substrates to meet their growth and energy requirements, enabling their application in diverse metal extraction scenarios (Buşilă et al., 2015). Moreover, microbes engaged in metal extraction showcase a remarkable affinity for specific metals. This selectivity is of utmost importance in achieving targeted metal recovery without the unwanted co-extraction of other elements.

Through the process of bioleaching, these exceptional microbes release organic acids or other compounds that aid in the solubilization of metals from ores or minerals. Their remarkable ability to selectively dissolve target metals greatly contributes to the efficiency and precision of metal extraction processes. Additionally, certain metal-extracting microbes possess the unique capability to immobilize or sequester metals within their biomass. This advantageous property allows for the concentration of metals, thereby simplifying subsequent recovery processes. Microbial biomass, enriched with metals, can be further processed to extract and refine the desired metal content (Štefelová et al., 2017). Moreover, microbes employed in metal extraction often exhibit robust growth characteristics, enabling them to swiftly colonize and adapt to changing environmental conditions. This particular feature is of utmost importance in ensuring the scalability and practicality of microbial metal extraction processes on an industrial scale. In conclusion, the properties of microbes involved in metal extraction encompass unparalleled metabolic adaptability, selectivity for specific metals, the remarkable ability to solubilize and immobilize metals, and robust growth characteristics. These extraordinary attributes collectively position them as invaluable tools in the pursuit of environmentally friendly and economically viable approaches to metal recovery from various sources, ranging from ores and minerals to electronic waste and spent batteries.

Microbes play a critical and essential role in the biosorption process, harnessing their exceptional biological capabilities to facilitate the uptake and conversion of substances in a wide range of environments. One of the key benefits of utilizing microbes in biosorption is their remarkable metabolic flexibility. Microorganisms exhibit a vast array of metabolic pathways, enabling them to interact with and absorb various substances, including nutrients, metals, and organic compounds. In the realm of metal bio absorption, certain microbes possess the remarkable ability to secrete organic acids or other chelating compounds (Zheng et al., 2013). These substances effectively bind with metals, promoting the dissolution of metal ions from solid substrates such as ores or minerals. This process, known as bioleaching, significantly enhances the bio absorption of metals, rendering them more accessible for microbial uptake or subsequent recovery.

Microbes also play a vital and indispensable role in environmental bioremediation, where their bio absorption capabilities are utilized to remediate contaminated sites. Some microbes have the capacity to absorb and convert pollutants, such as hydrocarbons or heavy metals, into less harmful forms. This microbial detoxification process contributes to the restoration of ecosystems impacted by industrial activities or pollution. In the field of agriculture, certain microbes establish symbiotic relationships with plant roots, significantly enhancing nutrient bio absorption. For instance, mycorrhizal fungi can extend the reach of plant roots, facilitating the absorption of crucial nutrients like phosphorus and micronutrients. This symbiotic interaction between microbes and plants promotes plant growth and overall crop productivity. This feature is of utmost importance in ensuring the scalability and practicality of microbial metal extraction processes on an industrial scale (Pereira et al., 2020).

Furthermore, microbes are indispensable in wastewater treatment processes, where they actively participate in the bio absorption and biodegradation of pollutants.

Microorganisms efficiently absorb and metabolize organic compounds present in wastewater, thereby contributing to the purification of water before it is reintroduced into the environment. In the medical field, the utilization of microbes in bio absorption processes is evident in certain therapeutic applications. For example, probiotic bacteria are employed to optimize the bio absorption of nutrients in the human digestive system, thus promoting overall gut health. Similarly, the use of microbial fermentation in the production of pharmaceuticals exemplifies the targeted bio absorption of specific compounds for therapeutic purposes (Bigham et al., 2014).

The incorporation of microbes in bio absorption processes aligns seamlessly with sustainable and eco-friendly practices. Microbial bio absorption typically occurs under gentle and moderate conditions, thereby reducing the need for harsh chemicals or energy-intensive processes. Additionally, the selectivity of certain microbial species ensures that bio absorption specifically targets desired substances, effectively minimizing the co-absorption of unwanted contaminants. In essence, the utilization of microbes in bio absorption processes underscores the versatility and adaptability of these microorganisms in interacting with diverse substances across various domains, including environmental remediation, agriculture, industry, and healthcare. Their exceptional biological capabilities make them invaluable agents for sustainable and targeted bio absorption applications (Enteria et al., 2013).

Project Aim and Objectives

While numerous studies have been undertaken on the biosorption of heavy metals for the purpose of soil bioremediation or wastewater cleanups, only very few studies have included or focused on the biosorption of lithium, a rare metal that is in high global demand for “green” technology applications and can be extracted from oil well and municipal wastewaters. The main purpose of this project is to demonstrate lithium biosorption and develop a methodology for the use of microbes in the absorption, concentration and recovery of lithium from aqueous solutions.

The objectives are:

- Procure, grow and harvest three microbes: B.*subtilis,* B.*cereus’* and E. *coli’*.
- Demonstrate lithium adsorption and desorption in a controlled environment.
- Identify the bacteria species that has the full potential to absorb lithium from lithium-laced aqueous solutions.
- Establish how contact time affects bio absorption of lithium and select the best exposure time for maximum biosorption.
- Develop a procedure(s) for the recovery of adsorbed metals using acid and/or pH spiking.
- Demonstrate the ability of regenerated bacterial biomass to reabsorb and desorb lithium for multiple uses.

Project Scope

The proposed study will involve several experiments focused on developing procedures for the testing of selected bacterial species or strains for adsorption and desorption of lithium under controlled laboratory environments which can be adapted on a commercial basis for the extraction of lithium, from wastewater derived particularly from oil fields or oil wells. The scope of work will include bacterial culturing and identification of a species or strain with optimal metal ion extraction function as advised by an independent specialist and subject to repeated laboratory tests.

The ideal conditions for the growth of the bacteria will also be described, along with the conditions under which the metal ion extraction process will be performed. The amount of lithium ion extracted will then be measured and the total amounts computed. Imputations will then be performed to assess the economic suitability of the proposed approach for sourcing raw material for high-energy fuel production, along with computed costs at an industrial scale. This will allow for the determination of the feasibility of the proposed microorganism approach to mass metal ion extraction. The scope of work may be limited by the availability of laboratory resources such as access to instruments for the precise analysis of lithium by ICP-OES or lithium selective ion electrodes.

CHAPTER 2

LITERARY REVIEW

Biosorption of lithium by using microbes represents a unique and sustainable method for lithium extraction, amidst developing worries about the environmental impact and sustainability of conventional extraction techniques. Biosorption is an environmentally friendly and economical process for removal of metals, an alternative to conventional methods such as chemical precipitation, electrochemical processes, reverse osmosis, ion exchange processes, and physical adsorption (Velkova et al., 2018). This literature overview finds out the modern state of research on microbial extraction of lithium, focusing on its functionality advantages, disturbing situations, and future possibilities. The biosorption of lithium through microbial methods offers a promising means for sustainable lithium extraction, bearing in mind the increasing apprehensions concerning the ecological repercussions and lengthy-term sustainability of traditional extraction strategies. As the worldwide call for lithium continues to surge, driven by the usage of the burgeoning electric car marketplace and continuous usage of electric devices, there can be a pressing need to discover opportunity strategies that decrease environmental impact whilst ensuring a stable supply of this vital beneficial resource.

This literature evaluation endeavors to delve into the cutting-edge-day panorama of studies surrounding microbial extraction of lithium, elucidating its capability benefits, inherent annoying situations, and avenues for future improvement (Yang et al., 2016).

Microbial extraction of lithium represents a paradigm shift from the conventional means of extracting lithium to an eco-friendlier extracting methodology, capitalizing on the metabolic skills of microorganisms to selectively leach lithium from various petroleum wells, ores and brines. This method presents the ability to mitigate the ecological footprint related to traditional mining and processing operations. By harnessing the natural strategies of microbes, microbial extraction endeavors to restrict habitat disruption, pollution problems, water wastage, and health problems associated with conventional extraction techniques. Moreover, microbial extraction holds the promise of reducing greenhouse gas emissions (Wang et al., 2019).

One of the number one hurdles lies in optimizing microbial methods for performance, selectivity, and scalability. The complex interactions amongst microorganisms and lithium-containing substrates necessitate a deeper understanding of microbial frame shape, biochemistry, and ecology to enhance manner performance. Additionally, the improvement of robust microbial traces with extra applicable lithium-leaching and tolerance to environmental fluctuations is important for the implementation of microbial extraction on a business scale. Furthermore, the monetary viability of microbial extraction techniques relative to traditional techniques remains a subject of scrutiny, requiring careful assessment of capital and operational fees and marketplace dynamics (Velkowa et al., 2018}.

While microbial extraction of lithium gives several advantages, inclusive of environmental sustainability and reduced strength consumption, it additionally faces big traumatic situations that ought to be addressed for successful implementation on an enterprise scale (Lo et al., 2014). The problematic interactions among microorganisms and lithium-containing substrates require a deeper knowledge of microbial body structure, biochemistry, and ecology to validate technique performance. Researchers must clarify the mechanisms underlying microbial-mediated lithium solubilization and uptake, as well as pick out key elements influencing approach overall performance, which consist of pH, temperature, and nutrient availability. By gaining insights into microbial metabolism and conduct, scientists can develop strategies to optimize technique situations and improve lithium extraction (Tsuruta, 2005).

Moreover, the improvement of strong microbial lines with higher lithium-leaching competencies is vital for a successful implementation of microbial extraction for a large business scale. Current microbial traces may additionally lack the vital trends to effectively solubilize and extract lithium from ores or brines, especially in difficult environmental situations. Researchers need to sharpen their genetic engineering skills to enhance microbial performance by engineering microbial lines with tailored functionalities, scientists can enhance system performance and reliability, thereby overcoming one of the maximum critical boundaries to big adoption of microbial extraction strategies. Furthermore, the financial viability of microbial extraction strategies relative to standard strategies remains a topic of scrutiny and requires careful evaluation. While microbial methods provide functionality benefits in phrases of environmental sustainability and selectivity, their financial competitiveness is based upon different factors, such as capital and operational costs, marketplace dynamics, and regulatory issues. (Tsuruta, 2005)

There is he need to assess the feasibility of microbial extraction in comparison to traditional strategies, thinking of factors inclusive of preliminary investment, working costs, and ability sales streams (Sethurajan and Gaydardzhiev, 2021). Additionally, exploring synergies with other industries or rate-delivered merchandise derived from microbial extraction techniques may enhance the economic viability of microbial extraction and assist offset initial implementation charges. At the same time as microbial extraction of lithium holds notable promise as a sustainable and environmentally friendly opportunity to standard strategies, it isn't without its disturbing situations.

Optimizing microbial processes for performance, selectivity, and scalability, growing strong microbial lines, and comparing the financial feasibility of microbial extraction techniques are important steps in figuring out the complete capacity of this progressive method. By addressing these challenges and advancing our expertise of microbial interactions with lithium-containing substrates, researchers can pave the way for the big adoption of microbial extraction strategies and make contributions to sustainable useful resource control practices (Illa et al., 2020).

Looking in advance, there are several avenues for future research and development within the problem of microbial extraction of lithium. Continued interdisciplinary collaboration among microbiologists, biochemists, engineers, and environmental scientists is important to reinforce our expertise of microbial strategies and optimize extraction techniques. Novel biotechnological and engineering procedures, together with genetic engineering of microbial traces and the design of bioreactor systems, promises a reinforcing system performance and scalability.

Moreover, exploring synergistic interactions among microbial consortia and integrating microbial extraction into included biorefinery ideas may additionally free up new possibilities for sustainable useful resource recuperation (Deng et al., 2019). In the end, microbial extraction of lithium represents a promising and sustainable method to meet the growing call for lithium at the same time as addressing environmental issues related to traditional extraction techniques. By leveraging the inherent metabolic talents of microorganisms, microbial extraction offers a pathway to limit ecological effect, lessen electricity consumption, and promote beneficial aid performance. However, overcoming key worrying situations and advancing our information of microbial interactions with lithium-containing substrates are crucial for understanding the overall ability of microbial extraction as a likely alternative to standard extraction techniques (Deng et al., 2019).

Before delving into microbial extraction techniques, it is important to understand the conventional techniques hired in lithium extraction. Conventional strategies incorporated in mining and processing lithium-containing ores, which incorporate spodumene and lithium-rich brines. These strategies are well-hooked up but often entail complex techniques and big environmental concerns, including habitat destruction, water consumption, and energy-extensive operations. Before embarking on an exploration of microbial extraction strategies, it is important to understand traditional strategies carried out in lithium extraction, which is the backbone of the lithium enterprise. Conventional strategies predominantly revolve around the mining and next processing of lithium-containing ores, which embody several mineral deposits wealthy in lithium, which incorporate spodumene, petalite, and lepidolite, amongst other (Illa et al., 2020).

Additionally, lithium-wealthy brines, located more often than not in salt residences or salars in regions like Chile, Argentina, and Bolivia, constitute another massive supply of lithium for extraction functions (Rautela et al., 2023). Mining of lithium-containing ores normally includes a sequence of steps, starting with exploration and drilling to discover and get entry to lithium deposits. Once discovered, the soil is extracted using open pit or underground mining techniques depending on factors such as grade, strength and availability of minerals. Following extraction, the ores undergo beneficiation processes to pay attention to the lithium content material, as lithium is frequently present in low concentrations in the ores. Beneficiation techniques may additionally consist of crushing, grinding, flotation, and magnetic separation to split lithium-bearing minerals from gangue minerals.

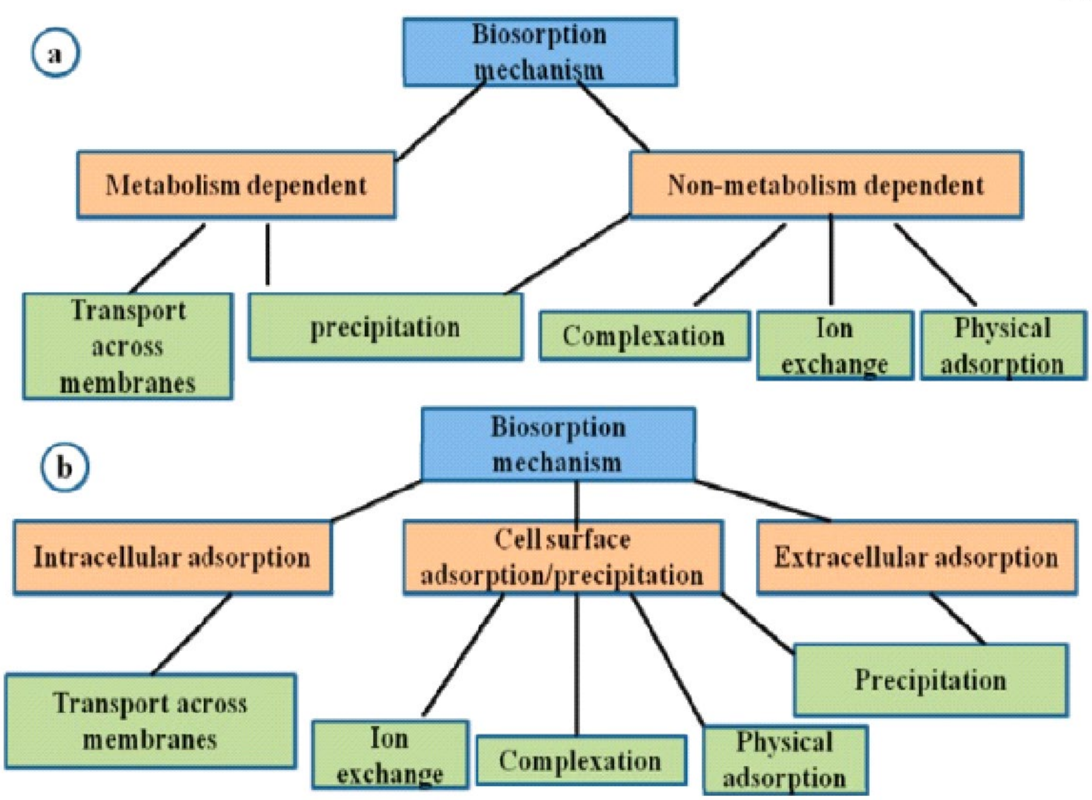

Figure 1: Mechanism of Biosorption Based on Dependence on Cell Metabolism, Location within the Cell where the Metal is Removed (Kure et al., 2018)

The mining of lithium-containing ores is a complicated process that includes several distinct tiers, each important for the extraction of lithium. Exploration and drilling mark the preliminary phase of the method, for the duration of which geologists and mining engineers survey ability sites to perceive and get entry to lithium deposits. This phase regularly involves good sized geological surveys, far off sensing techniques, and geophysical investigations to pinpoint areas with excessive lithium concentrations. Once suitable deposits are recognized, drilling operations are carried out to extract core samples and determine the fineness and the quantity of lithium-bearing ores gifted below the earth's surface (Xie et al., 2018).

These exploratory efforts are important for determining the feasibility of mining operations and optimizing useful resource extraction. Mining of lithium-containing ores is a multifaceted process that entails a series of meticulously orchestrated steps aimed at extracting this valuable resource. The journey commences with exploration and drilling endeavors, integral for pinpointing potential lithium deposits. Geologists and mining engineers collaborate closely during this phase, employing an array of sophisticated techniques such as geological surveys, remote sensing technologies, and geophysical investigations. These efforts are essential for identifying promising sites with elevated lithium concentrations. Through meticulous examination of core samples extracted via drilling operations, experts assess the quality and quantity of lithium-bearing ores lurking beneath the earth's surface. Such exploratory initiatives serve as the bedrock for gauging the feasibility of subsequent mining operations, crucial for optimizing resource extraction (Wang et al., 2019).

Upon successful identification of viable lithium deposits, the extraction process unfolds, encompassing both open-pit and underground mining methodologies. Selection between these techniques hinges upon various factors including ore grade, structural integrity, and mineral availability. Open-pit mining, characterized by its cost-effectiveness and efficiency in accessing shallow deposits, is often preferred when feasible. Conversely, underground mining is pursued when deposits delve deeper into the earth's crust, necessitating specialized excavation techniques to ensure safe and efficient extraction. Regardless of the method employed, meticulous planning and execution are imperative to safeguard both personnel and environmental integrity throughout the extraction phase.

With ores procured from the mining operations, the beneficiation stage ensues, a critical juncture aimed at concentrating the lithium content. Given that lithium is frequently dispersed in low concentrations within the ores, beneficiation techniques play a pivotal role in enhancing its concentration for subsequent processing (Xin et al., 2009). Crushing and grinding operations pulverize the extracted ores into finer particles, facilitating the separation of lithium-bearing minerals from their gangue counterparts. Subsequent flotation and magnetic separation processes further refine the ore, isolating valuable lithium constituents from undesirable impurities. The efficacy of these beneficiation techniques hinges upon their ability to selectively extract lithium-rich minerals, thereby enhancing overall extraction efficiency.

Throughout the mining process, stringent environmental protocols are adhered to mitigate ecological impact and ensure sustainable resource utilization. Comprehensive environmental impact assessments are conducted prior to mining commencement to identify potential risks and develop mitigation strategies. Measures such as land rehabilitation and water management initiatives are implemented to mitigate habitat disruption and safeguard natural ecosystems. Additionally, efforts are directed towards minimizing energy consumption and greenhouse gas emissions associated with mining activities, underscoring the industry's commitment to environmental stewardship (Lo et al., 2014).

In essence, the mining of lithium-containing ores epitomizes a harmonious interplay of scientific acumen, technological innovation, and environmental conscientiousness. From initial exploration endeavors to the meticulous execution of extraction and beneficiation processes, each stage of the journey is meticulously orchestrated to unlock the full potential of this indispensable resource. As the global demand for lithium continues to surge in tandem with burgeoning advancements in renewable energy and electric mobility, responsible mining practices remain paramount in ensuring a sustainable and equitable future for generations to come.

Following the exploration section, the real extraction of lithium-containing ores begins, usually utilizing either open-pit or underground mining techniques. The desire among those strategies relies upon different factors, which include the ore grade, intensity of the deposit, geological traits, and environmental concerns. Open-pit mining, the maximum commonplace approach for extracting lithium ores close to the surface, entails the excavation of large pits or quarries to access shallow deposits of lithium-bearing minerals. This method is enormously fee-effective and allows for green extraction of big quantities of ore. Conversely, underground mining is hired to access deeper deposits positioned underneath the earth's floor. This technique includes the development of tunnels, shafts, and galleries to reap the ore body, requiring specialized devices and safety precautions due to the underground running situations (Lo et al., 2014). Once the lithium-containing ores are extracted from the earth, they undergo beneficiation strategies to concentrate the lithium content material and take away impurities. Lithium is regularly found in low concentrations in the ores, necessitating beneficiation techniques to increase its recognition and facilitate in addition processing (Lo et al., 2014).

Beneficiation techniques range from relying on the composition of the ores and might encompass mechanical procedures along with crushing, grinding, and screening to lessen the ore duration and unencumbered lithium-bearing minerals from the encircling gangue minerals. Additionally, bodily separation strategies together with flotation and magnetic separation are hired to selectively separate lithium-bearing minerals from the gangue minerals based on their bodily houses, together with density, magnetic susceptibility, and surface chemistry.

Overall, the mining of lithium-containing ores involves a chain of interconnected steps, from exploration and drilling to extraction and beneficiation. Each stage of the system performs an important function in obtaining super lithium concentrates appropriate for further processing into lithium compounds or products. While advancements in mining technology and processing techniques have superior the performance and sustainability of lithium mining operations, ongoing research and innovation are had to deal with environmental worries, optimize useful resource utilization, and make certain the accountable extraction of lithium assets for destiny generations (Mishra et al., 2008).

After beneficiation, the centered lithium ores undergo further processing to extract lithium in a usable form, usually as lithium carbonate or lithium hydroxide. This processing typically includes roasting or calcination of the centered ores to remove impurities and convert lithium-bearing minerals into water-soluble lithium compounds. Acid leaching or alkaline extraction techniques are then hired to dissolve lithium from the roasted ores, observed with the aid of precipitation and purification steps to attain lithium carbonate or lithium hydroxide. While conventional lithium extraction techniques had been instrumental in assembly global lithium call for, they will be no longer without environmental concerns and logistical demanding situations. Habitat destruction, especially in ecologically sensitive regions hosting lithium deposits, is a massive environmental outcome of mining sports activities. Open-pit mining operations can cause the clearance of vegetation, disruption of flora and fauna habitats, and alteration of landscape topography. Similarly, underground mining might also result in subsidence and land instability, posing risks to surrounding ecosystems and communities (Rezza et al., 2001). Moreover, the water-intensive nature of lithium extraction, particularly inside the case of lithium-wealthy brine deposits, will increase concerns concerning water consumption and capacity effects on local water resources.

Extraction of lithium from brines frequently includes pumping large volumes of brine to the floor and next evaporation in open ponds, necessitating massive water usage. This can strain water factors in arid regions and exacerbate competition for water sources, in particular areas already facing water scarcity or competing needs for water utilization (Illa et al., 2020). Furthermore, the power-extensive nature of traditional lithium extraction methods, in particular all through ore beneficiation and chemical processing stages, contributes to carbon emissions and reliance on non-renewable electricity resources. High electricity consumption is not handiest to the environmental footprint but additionally raises operational expenses and vulnerabilities to energy charge fluctuations. In stop, whilst traditional techniques of lithium extraction have performed a pivotal role in meeting international lithium call for, they are accompanied by making use of massive environmental worries and logistical challenges. Habitat destruction, water intake, and strength-extensive operations are most of the key problems related to traditional extraction techniques. As such, there may be a development critical to explore possibility techniques to lithium extraction, which include microbial extraction, which offer the capability for decreased environmental impact and improved sustainability (Rautela et al., 2023).

The exploration of opportunity techniques for lithium extraction stems from numerous environmental and sustainability issues. Conventional extraction strategies may want to have profound ecological influences, which consist of habitat destruction, water utilization, and strength consumption. The transition towards extra sustainable extraction methods aligns with global efforts to mitigate environmental degradation, lessen carbon emissions, and sell circular financial device concepts. The quest for possible techniques of lithium extraction arises from a confluence of environmental and sustainability imperatives, underpinned with the aid of mounting issues over the ecological footprint of traditional extraction practices (Deng et al., 2019). Conventional extraction techniques, characterized through their reliance on mining and chemical processing strategies, impose sizable environmental burdens, in phrases of habitat destruction, water consumption, and strength utilization. The ecological influences of such practices extend past the immediate area of mining, encompassing broader ecosystems and water assets, thereby necessitating a reevaluation of cutting-edge extraction paradigms.

Habitat destruction emerges as a distinguished environmental end result of conventional lithium extraction, as mining regularly entails the clearing of plants, alteration of landscapes, and disruption of flowers and fauna habitats. The conversion of natural habitats into commercial landscapes can bring about biodiversity loss and ecological fragmentation, posing dangers to endemic species and surroundings resilience. Water usage represents any other crucial difficulty associated with standard lithium extraction techniques, particularly inside the context of lithium-rich brine deposits (Xie et al., 2018). The extraction of lithium from brines normally includes the pumping of large volumes of brine to the floor and next evaporation in open ponds, necessitating vast quantities of water. This water-extensive method does not most effectively strain local water assets; however it also raises problems over water great and environment integrity. In regions already grappling with water scarcity or competing wishes for water utilization, which include arid or semi-arid environments, the extraction of lithium from brines can exacerbate water stress and heighten tensions over water allocation. Water usage is certainly an important difficulty associated with conventional lithium extraction techniques, in particular concerning lithium-rich brine deposits. The manner of extracting lithium from brines normally entails the pumping of massive volumes of brine to the ground, located through evaporation in open ponds to pay interest to the lithium content material (Xie et al., 2018)

However, this method necessitates great portions of water, elevating several environmental and sustainability annoying situations. One of the number one troubles is the stress located on neighborhood water property, especially in regions wherein water availability is limited. In arid or semi-arid environments, wherein water scarcity is already an urgent problem, the extraction of lithium from brines can exacerbate water stress and heighten tensions over water allocation. Competing demands for water utilization, which encompass agriculture, municipal supply, and environment protection, similarly compound the challenges related to lithium extraction, leading to conflicts over water get proper of entry to and distribution (Matejczy et al., 2018).

Moreover, the extraction of lithium from brines could have detrimental influences on water satisfaction and environment integrity. The pumping and evaporation of brines can disrupt herbal hydrological cycles and modify the composition of neighborhood water bodies, potentially leading to salinization, contamination, and habitat degradation. High ranges of salt and different contaminants found in brines can impair water first-rate, rendering it improper for human consumption, agricultural irrigation, or aquatic habitats. Additionally, the discharge of brine effluents into surrounding water bodies can disrupt aquatic ecosystems, affecting fish and flora and fauna populations and compromising universal atmosphere fitness (González-Gil, Izquierdo, Marcos and Palacios, 2011).

These environmental impacts highlight the need for sustainable water management practices and mitigation measures to limit the adverse effects of lithium extraction on water sources and ecosystems. Furthermore, the extraction of lithium from brines presents unique environmental challenges that necessitate careful consideration and mitigation strategies. Unlike traditional mining methods, which involve excavating solid ores, lithium extraction from brines involves the pumping and evaporation of saline groundwater. While brine extraction offers certain advantages, such as lower production costs and a smaller physical footprint compared to conventional mining, it also poses significant risks to water quality and environmental integrity.

The pumping of brines from underground aquifers can disrupt natural hydrological cycles, leading to changes in groundwater levels and flow patterns. This alteration can have far-reaching consequences, including the depletion of local water reserves and the destabilization of fragile ecosystems reliant on groundwater resources (Xu et al., 2018). Moreover, due to the extensive hydrological impacts, the composition of brines contains high levels of salts and other contaminants that can degrade water quality and compromise ecosystem health.

The discharge of brine effluents into surface water bodies can lead to salinization, a process whereby the accumulation of salts renders water unfit for human consumption, agricultural use, or aquatic life. Evaporation of brines in large-scale lithium extraction operations can exacerbate these effects, further intensifying water scarcity and habitat disruption in surrounding areas. The elevated salinity levels can disrupt the balance of aquatic ecosystems, leading to the decline of native species and the proliferation of salt-tolerant organisms, further destabilizing the ecological equilibrium (Shim, Jin, Seo, Lee and Kim, 2011).

Furthermore, the discharge of brine effluents can introduce toxic elements and heavy metals into aquatic environments, posing risks to fish and wildlife populations and threatening human health. Contaminants such as lithium, magnesium, and boron, which are commonly present in brines, can bioaccumulate in aquatic organisms, potentially leading to adverse effects on reproduction, growth, and survival. These ecological disruptions can have cascading effects throughout the food chain, ultimately compromising the overall health and resilience of aquatic ecosystems. To address these environmental concerns, proactive measures and sustainable

Implementing effective monitoring and mitigation strategies can help minimize the impacts of lithium extraction on water quality and ecosystem integrity (Ye et al., 2019). This may involve the development of innovative technologies for brine treatment and disposal, as well as the implementation of stringent regulatory frameworks to ensure compliance with environmental standards. Furthermore, stakeholder engagement and community involvement are crucial for fostering transparency and accountability in the lithium extraction process. By involving local communities, indigenous groups, and environmental organizations in decision-making processes, industry stakeholders can better identify and address the concerns and priorities of affected communities. This collaborative approach can help build trust and foster sustainable development practices that prioritize environmental conservation and social responsibility (Yoon et al., 2005).

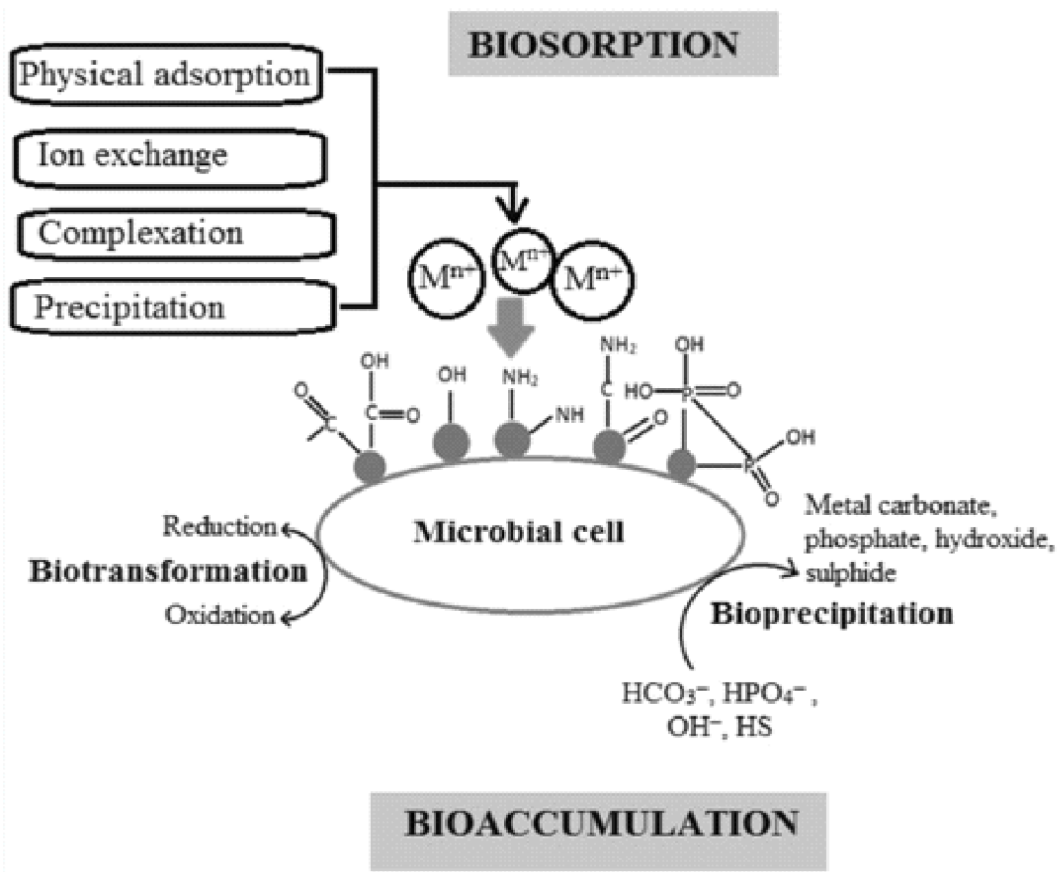

Figure 2: Flow Chart Showing Relationship Between Biosorption and Bioaccumulation (Hansda et al., 201)

In conclusion, the extraction of lithium from brines presents both opportunities and challenges for meeting the growing demand for this critical resource. While brine extraction offers certain advantages in terms of cost and efficiency, it also poses significant risks to water quality and ecosystem health. By implementing proactive mitigation measures and sustainable water management practices, stakeholders can minimize the environmental impacts of lithium extraction and ensure the responsible stewardship of natural resources for future generations.

Furthermore, the water-intensive nature of conventional lithium extraction strategies increases issues over the long-term sustainability of useful resource extraction and control. As global demand for lithium maintains an upward push, driven by the developing adoption of electrical automobiles and renewable electricity technology, the pressure on water sources associated with lithium extraction is predicted to boom considerably. In areas where water shortage is already an essential trouble, such as the lithium-rich salars of South America's "lithium triangle," sustainable water management strategies are urgently needed to make certain the accountable and equitable use of water resources. This may also involve enforcing water-saving technology, recycling and reuse of procedure water, and adopting opportunity extraction methods that decrease water usage and environmental effect. In the end, water utilization represents a giant subject related to conventional lithium extraction methods, especially within the context of lithium-rich brine deposits (Yun & Volesky, 2003).

The extraction of lithium from brines requires huge volumes of water, straining neighborhood water resources, compromising water fine, and threatening surroundings integrity. In regions already dealing with water scarcity and competing needs for water utilization, the extraction of lithium can exacerbate water strain and exacerbate tensions over water allocation. Sustainable water control practices, alongside technological innovation and opportunity extraction methods are essential for mitigating the environmental influences of lithium extraction and ensuring the long-term sustainability of lithium sources (Wang, 2018).

From ore beneficiation to chemical processing, those operations require big amounts of energy, predominantly derived from fossil fuels, thereby exacerbating climate trade and environmental degradation. The transition in the direction of extra sustainable extraction techniques, characterized through lower electricity necessities and reliance on renewable energy resources, represents a vital step toward decarbonizing the lithium supply chain and advancing global efforts to mitigate weather change. In response to those environmentally demanding situations, there is a growing popularity of the want to promote circular economy standards within the extraction and utilization of lithium. A circular economy technique emphasizes aid efficiency, waste minimization, and the valorization of secondary assets, thereby reducing reliance on virgin materials and mitigating environmental effects.

By prioritizing the recycling and restoration of lithium from end-of-existence products, consisting of spent lithium-ion batteries, and exploring alternative extraction methods, which includes microbial extraction, the lithium industry can circulate closer to a greater sustainable and resilient destiny (Cubillos et al., 2018). Furthermore, the exploration of alternative strategies for lithium extraction is pushed making use of a developing consciousness of the environmental and sustainability demanding situations related to standard extraction practices. Habitat destruction, water usage, and energy intake are most of the key environmental worries motivating the transition in the direction of greater sustainable extraction strategies (Roy, 2021). Microbial extraction of lithium offers more promising opportunities than conventional methods, the metabolism of specific microorganisms is used to select lithium from minerals or salts This method has many advantages, including environmental sustainability, overall power basic performance and minimum ecological impact. Biological processes operate under normal conditions, reducing energy consumption and greenhouse gas emissions by permitting the selective recovery of lithium, microbial extraction offers a more sustainable and environmentally friendly approach to useful resource extraction, aligning with broader dreams of useful resource conservation and environmental safety.

Moreover, the selectivity of microbial extraction streamlines downstream processing and decreases the complexity of ore beneficiation and purification. Conventional extraction strategies frequently require great processing steps to separate lithium from other metals and minerals present within the ore or brine, leading to improved energy consumption, chemical usage, and environmental impact. In contrast, microbial extraction can lead to a more sustainable means of Lithium extraction, facilitating the direct solubilization and recuperation of lithium ions without the need for complex separation techniques. This simplification of the extraction method no longer only improves operational efficiency, however, also lowers fees and reduces the environmental footprint of lithium extraction operations (Evers et al., 2012). Additionally, by way of minimizing the use of chemicals and electricity-extensive processing steps, microbial extraction contributes to the general sustainability and eco-friendliness of lithium manufacturing.

Furthermore, the selectivity of microbial extraction techniques offers opportunities for useful resource healing and valorization. In addition to targeting lithium, microbial approaches can be engineered to extract and get better different precious metals or minerals present in the ore or brine, along with cobalt, nickel, and rare earth elements. By harnessing the metabolic abilities of microorganisms, researchers can increase biotechnological solutions for the sustainable extraction and utilization of more than one resource from complex geological formations. This multi-steel restoration technique no longer most effectively maximizes useful resource usage and price advent but also reduces the environmental effect of mining operations with the aid of minimizing waste technology and aid depletion (Buşilă et al., 2015). Moreover, by recovering extra metals and minerals, microbial extraction complements the monetary viability of lithium production and promotes the development of included mining and processing systems that optimize aid efficiency and sustainability. In the end, microbial extraction gives inherent selectivity, permitting centered recuperation of lithium without co-extracting unwanted impurities.

Despite the potential, microbial removal of lithium faces many challenges and obstacles that need to be overcome to achieve commercial application These include providing microbial pathways well and able to grow under stressful conditions, knowledge of microbial interactions with lithium-containing substrates in addition to these, the economic viability of microbial removal techniques in standard systems that examined requires similar analysis. Achieving essential manners is critical to maximize lithium recuperation costs and make sure of the financial viability of microbial extraction strategies. Additionally, scaling up microbial approaches from laboratory-scale experiments to business-scale operations gives logistical demanding situations that have to be addressed, which includes engineering concerns, manner automation, and cost-effectiveness.

Understanding microbial interactions with lithium-containing substrates is any other key assignment in microbial extraction of lithium. While certain microorganisms have been diagnosed for his or her ability to leach lithium from ores or brines, the mechanisms underlying microbial lithium solubilization and uptake are nonetheless not completely understood. Further research is needed to elucidate the biochemical pathways and metabolic tactics worried in microbial-mediated lithium extraction, that allows you to inform the improvement of greater efficient and effective microbial extraction techniques (Tao et al., 2014). Additionally, exploring the variety of microbial communities in lithium-rich environments and their potential packages in lithium extraction might also find novel microbial candidates with greater lithium-leaching capabilities.

Microbial extraction of lithium gives a promising road for assembly the growing demand for this essential detail in various industries, especially in the realm of renewable electricity technology. However, despite exceptional improvements, knowledge of microbial interactions with lithium-containing substrates remains a big assignment. One of the primary hurdles lies in elucidating the complicated mechanisms that govern microbial-mediated lithium solubilization and uptake. While excellent microorganisms have tested the capability to leach lithium from ores or brines, the underlying biochemical pathways and metabolic methods concerned on this phenomenon are not but truly understood.

In addition to elucidating the crucial mechanisms of microbial-mediated lithium extraction, practical problems such as way optimization and scalability should be addressed. Developing robust bioprocess engineering strategies that integrate microbial body structure, substrate chemistry, and reactor design is crucial for translating laboratory-scale findings into feasible commercial applications (Tao et al., 2014). Furthermore, interdisciplinary collaborations between microbiologists, chemists, engineers, and agency stakeholders are vital for accelerating the improvement and deployment of microbial based totally lithium extraction generation. In conclusion, while exceptional strides were made in harnessing microorganisms for lithium extraction, numerous demanding situations and possibilities lie ahead. By advancing our understanding of microbial interactions with lithium-containing substrates, elucidating biochemical pathways, exploring microbial diversity, and integrating bioprocess engineering concepts, we will pave the way for the improvement of sustainable and inexperienced microbial-based total tactics for lithium healing.

Such endeavors are important for assembling the developing call for lithium at the same time as minimizing environmental effects and promoting a transition toward a renewable electricity future (Jegan Roy et al., 2021). Mitigating capability environmental dangers associated with microbial activities is also an essential attention within the development of microbial extraction techniques. While microbial procedures carry out below moderate situations in assessment to standard extraction techniques, there's a hazard of unintentional environmental effects, which incorporates the discharge of dangerous byproducts or the introduction of non-nearby microorganisms into natural ecosystems.

At the same time as microbial extraction of lithium holds great promise as a sustainable and environmentally tremendous technique to useful resource extraction, addressing the challenges and barriers stated above is essential for a successful implementation on an enterprise scale. By optimizing microbial techniques for performance and scalability, knowledge microbial interactions with lithium-containing substrates, mitigating functionality environmental dangers, and making sure monetary viability, microbial extraction techniques can play a critical role in advancing sustainable useful resource control practices and reducing the environmental footprint of lithium extraction operations (Hashimoto et al., 2014). Continued studies, innovation, and collaboration across disciplines is probably key to overcoming those disturbing conditions and knowing the complete capability of microbial extraction of lithium.

Future studies in microbial extraction of lithium need attention on addressing the challenges whilst exploring opportunities to enhance process performance, selectivity, and sustainability. This may also involve the improvement of novel microbial lines with optimized lithium-leaching abilities, the integration of biotechnological and engineering methods to scale up microbial methods, and the research of synergistic interactions among microbial consortia for superior lithium extraction. Moreover, interdisciplinary collaboration amongst microbiologists, biochemists, engineers, and environmental scientists can be critical for advancing the world and identifying the entire capability of microbial extraction of lithium. The future of research in microbial extraction of lithium holds full-size promise, with a focal point on addressing gift demanding situations at the same time as exploring avenues to beautify machine performance, selectivity, and sustainability.

One avenue for advancement entails the development of novel microbial lines with optimized lithium-leaching competencies. By leveraging advances in biotechnology and genetic engineering, researchers can engineer microorganisms to decorate their capacity to solubilize and extract lithium from ores or brines. This method also can moreover contain modifying microbial metabolic pathways, improving enzyme sports activities, or introducing novel biosynthetic pathways to decorate lithium restoration costs and technique performance (Wu et al., 2019). Additionally, screening microbial variety from numerous environments may additionally find natural lines with inherent lithium-leaching competencies, providing precious insights into the capability for microbial-based totally absolutely techniques in lithium extraction.

The future of studies in microbial extraction of lithium holds great promise, marked with the resource of a concerted attempt to address present demanding conditions at the same time as exploring avenues to beautify technique performance, selectivity, and sustainability. One promising avenue for advancement lies in the development of novel microbial strains engineered with optimized lithium-leaching competencies. Leveraging upgrades in biotechnology and genetic engineering, researchers can tailor microorganisms to enhance their ability to solubilize and extract lithium from numerous assets, which incorporate ores and brines. This approach may additionally entail the manipulation of microbial metabolic pathways, augmentation of enzyme sports, or advent of novel biosynthetic pathways to expand lithium recuperation charges and system efficiency (Zeng et al., 2013).

Moreover, the screening of microbial range sourced from diverse environments provides some other promising avenue for discovery. By tapping into the vast reservoir of microbial diversity present in extraordinary ecosystems, researchers can find herbal strains endowed with inherent lithium-leaching capabilities. The exploration of microbial groups inhabiting lithium-rich environments holds the ability to yield precious insights into the variety of metabolic techniques hired by way of microorganisms to interact with lithium-containing substrates. Through systematic screening and characterization efforts, researchers can discover microbial applicants with desirable developments for superior lithium extraction, paving the way for the development of extra sustainable and efficient microbial-primarily based tactics in lithium recuperation. Integration of biotechnological and engineering approaches is some other crucial thing of destiny studies in microbial extraction of lithium. Scaling up microbial strategies from laboratory-scale experiments to commercial-scale operations requires revolutionary engineering solutions and procedure optimization.

By integrating bioreactor format, technique automation, and monitoring technology, researchers can broaden green and price-effective strategies for big-scale microbial extraction of lithium. Moreover, advances in bioprocess engineering, which include non-stop float systems and immobilized cell technologies, might also in addition decorate manners of efficiency and productivity, paving the way for commercialization of microbial extraction techniques. Exploration of synergistic interactions among microbial consortia provides some other thrilling possibilities for reinforcing lithium extraction performance (Zhang et al., 2012).

Microbial communities in herbal environments often showcase complicated interactions and cooperative behaviors, which can be harnessed to enhance lithium recuperation rates and selectivity. By analyzing microbial consortia from lithium-rich environments and elucidating their metabolic interactions, researchers can discover key microbial gamers and optimize their interactions to decorate lithium extraction overall performance. Moreover, engineering synthetic microbial consortia with tailored functionalities may additionally allow specific manipulation over lithium solubilization and uptake, further improving overall performance and sustainability.

Interdisciplinary collaboration among microbiologists, biochemists, engineers, and environmental scientists can be vital for advancing the world of microbial extraction of lithium. By combining knowledge from numerous disciplines, researchers can address complex disturbing situations and develop holistic answers that integrate natural, chemical, and engineering ideas. Collaborative research efforts can boost progress in information microbial interactions with lithium-containing substrates, optimizing process situations, and mitigating environmental risks associated with microbial activities. Moreover, interdisciplinary collaboration fosters innovation and creativity, important to the improvement of novel processes and technologies for sustainable lithium extraction (Lokhande et al., 2022)

In conclusion, destiny studies in microbial extraction of lithium have to pay attention to addressing challenges at the same time as exploring opportunities to decorate technique, overall performance, selectivity, and sustainability. This may additionally contain the improvement of novel microbial strains, integration of biotechnological and engineering approaches, exploration of synergistic microbial interactions, and interdisciplinary collaboration (Lokhande et al., 2022). By advancing our expertise in microbial tactics and leveraging biotechnological and engineering improvements, researchers can unencumber the total capacity of microbial extraction methods for sustainable and environmentally friendly lithium extraction.

In the end, microbial extraction of lithium holds great promise as a sustainable and environmentally friendly alternative to conventional extraction methods. By harnessing the metabolic sports of microorganisms, microbial tactics provide a pathway to mitigate environmental effects, lessen power consumption, and promote the circular economy. However, addressing key challenges and advancing our know-how of microbial interactions with lithium-containing substrates are vital for realizing the full capability of this innovative technique. Future studies ought to put more attention on optimizing microbial approaches, enhancing system efficiency and selectivity, and exploring interdisciplinary collaborations to create innovation in the subject of microbial extraction of lithium (Kaikhosrov et al., 2021).

The potential for microbial extraction of lithium as the first sustainable and environmentally friendly opportunity in conventional systems is certainly high. Biological approaches using precise microbial metabolism offer a promising approach to reduce the environmental impact associated with conventional extraction methods What unlike conventional systems, which typically involve habitat degradation, water use, and power-intensive operations, microbial extraction operates under suboptimal conditions This freedom to they offer an outcry for environmental impact It fits with broader dreams of sustainability and the environment, making microbial extraction an attractive option meeting the growing call for lithium, while reducing its ecological footprint.

Additionally, microbial extraction of lithium has the potential to reduce energy consumption, any other important factor in environmental sustainability. Conventional extraction techniques regularly require significant quantities of energy for ore processing, chemical treatment, and transportation, contributing to carbon emissions and exacerbating weather change. In contrast, microbial techniques perform below pretty low energy inputs, as microorganisms utilize their metabolic pathways to solubilize and extract lithium from ores or brines. This power-green approach is the most effective in reducing greenhouse gas emissions but moreover lowers operational prices, improving the economic viability of microbial extraction techniques (Kaikhosrov et al., 2021).

Moreover, microbial extraction of lithium aligns with the ideas of the round economic system by promoting resource recuperation and reuse. As the demand for lithium continues to upward push, there is a growing desire to discover alternative resources and extraction methods to ensure a sustainable supply chain. Microbial extraction offers an answer by permitting the healing of lithium from low-grade ores, waste materials, and spent batteries, thereby decreasing the reliance on virgin sources and minimizing waste technology. By getting better treasured resources from secondary sources, microbial extraction contributes to the greenhouse assets and the discount of environmental effect, advancing the desires of the round economy. However, to absolutely realize the capacity of microbial extraction of lithium, it is miles critical to deal with key demanding situations and advance our know-how of microbial interactions with lithium-containing substrates.

One task is optimizing microbial processes for efficiency and selectivity, as factors including pH, temperature, nutrient availability, and microbial community composition can drastically have an impact on lithium extraction quotes. Additionally, know-how the mechanisms underlying microbial-mediated lithium solubilization and uptake is important for developing tailored microbial lines and optimizing procedure situations (Kinoshita, 2019). Collaborative research efforts concerning microbiologists, biochemists, engineers, and environmental scientists could be vital for advancing our expertise of microbial methods and developing revolutionary answers to triumph over these demanding situations. In the end, microbial extraction of lithium holds mammoth promise as a sustainable and environmentally pleasant alternative to traditional strategies (Kinoshita, 2019).

By harnessing the metabolic activities of microorganisms, microbial techniques offer a pathway to mitigate environmental effects, lessen strength consumption, and sell the circular economic system. However, addressing key challenges and advancing our know-how of microbial interactions with lithium-containing substrates are critical for realizing the whole capacity of this revolutionary approach. Future research efforts must focus on optimizing microbial methods, enhancing procedure efficiency and selectivity, and exploring interdisciplinary collaborations to power innovation in the area of microbial extraction of lithium (Vanitha et al., 2013).

CHAPTER 3

DESIGN OF THE STUDY

The study was conducted by first procuring all the necessary materials needed for the experiments followed by experiments based on well established procedures.

Materials

Microorganisms

Escherichia *coli*

Bacillus *subtilis*

Bacillus *cereu*s

Reagents

LiCl 99.9%, 100 g

HNO3 (Conc)

100 g of agar growth media powder

100 g of LB broth powder

Yeast extract

Agar powder

Glucose

Laboratory Equipment

Petri dishes for solid media

Hot Plate

Inoculating loop

Orbital shaker

Autoclave

Deep Freezer

Incubation heating lamp with temperature control

Bunsen Burner for aseptic technique

Centrifuge and compatible tubes

Microfilters

Precision scale

TDS/pH/Temperature meter

Spray bottles

Test tubes 50 mLand Rack

Nitrile gloves

Disposable transfer pipettes x100

0.5 L flasks

1 L flask

1 L and 250 mL graduated cylinders

Glass beakers (50, 100, 150, 200 ml)

Plastic cups x100

Lab surface (table or desk)

Heat resistant gloves

Permanent and label marker

Orbital shaker

Incubation heating lamp with temperature control

Inductively Coupled Plasma-Optical Emission Spectroscopy (ICMP-OES)

Procedures

The bacteria species provided were Escherichia *coli,* Bacillus *subtilis*, Bacillus *cereu*s and Bacillus *megaterium*, Escherichia *coli* was all purchased from ATCC in a freeze-dried form. while the other three were obtained through the assistance of my Project Advisor. These bacteria species were selected based on research and their ability to bio absorb metals. 100g of lithium chloride (LiCl) was also purchased from Amazon for the preparation of standard solutions. All other equipment like petri dishes, inoculating loop, orbital shaker, hot plate, water bath, double distilled water and TDS meter are already available in the laboratory.

The materials to be used were carefully and neatly unpackaged and arranged in the laboratory before the commencement of laboratory activity. Inoculation equipment such as spatulas, petri dishes and inoculating loops were autoclaved at 15 psi for 15 minutes. Laboratory surfaces were sprayed down and cleaned with 70% alcohol and lab instruments were sterilized with the autoclave~~d~~. The combined Temperature/TDS/pH Meter was calibrated and always zeroed and cleaned in distilled water prior to and after every sample measurement. For lithium analysis, the Inductively Coupled Plasma - Optical Emission Spectroscopy (ICP-OES) equipment in the laboratory of Dr Gao at the University of Houston was used. Linear calibration curves for lithium were prepared by Dr. Gao using a range of standard solutions.

Rehydration of Microbial Biomass

Rehydration is the process of restoring lost water to bacterial biomass. To rehydrate the bacteria procured from the American Type Culture Collection (ATCC) supply, 1 ml of LB broth is aseptically added into the ampoule bottles containing *Escherichia coli,* (the preferred bacteria species used in this project) t *Bacillus subtills and* Bacillus *cereus and* stirred on a vortex for 10 minutes. 5ml is transferred from the ampoule bottle into a test tube containing 3 ml of LB broth and allowed to sit for 3 hours to rehydrate the bacteria in all the 3 bottles.

Preparation of Lithium Solutions

The appropriate quantity of lithium chloride salt (LiCl) was carefully measured out and diluted with 500 ml of distilled water to make a 500-ppm stock solution. From this stock solution, standards for 50 ppm and 150 ppm were prepared as appropriate by further dilution with distilled water or broth solution. More details are presented in a subsequent section on lithium biosorption experiments.

Culture Preparation

In cultivating microbial biomass, the culture medium used was Lysogeny Broth (LB) which is a nutritionally rich medium most commonly utilized for the growth of bacteria.

<u>Lysogeny Broth (LB) Medium</u>

The preparation was as follows: 12.5 g of LB powder was added to 500 mL of distilled water in a flask, mixed, and heated up in the autoclave for 30 minutes to sterilize. The directions on the box were followed in adjusting the pH if necessary. Autoclave was opened as pressure dropped to 1 atm. They are allowed to cool down and kept aside until when needed.

Inoculation and Culturing

Luria broth agar (LB) is a multi-purpose rich medium known to grow both gram positive and gram-negative bacteria. It contains 5g of yeast extract, 10g of peptone and 10g of sodium chloride and 12g of agar. To prepare the medium, 37.5g of (LB) agar was measured into 1000 ml of distilled water. It was allowed to dissolve, and then autoclaved at 12^{o}C for 30 minutes. Once properly autoclaved, the petri dishes were labeled with the name of the appropriate bacteria then poured aseptically into 21 petri dishes and allowed to cool down. The inoculating loop was sterilized by flaming it until it was red hot. Bacteria was transferred from the 3 ml test tube to the agar plate by streaking on the agar plates. Incubate at 37°C for 24 hours to allow the biomass to grow. Repeat the same process for E *col*i, Bacillus *subtilis,* and Bacillus *cereus*. The remaining rehydrated bacteria in the test tube was preserved by adding 30% glycerol stock and stored in a -70 freezer.

Overnight Sub-culturing

The LB broth prepared earlier was used to subculture the bacteria. The *E. coli* biomass grown for 24 hours on LB agar plate was scraped aseptically into 100 ml of LB broth, scraping the biomass of *Bacillus subtilis* grown for 24 hours into another 100ml. Repeat the same process for *Bacillus cereus* and incubate the 3 bottles at 37°C for 24 hours. The next day check the optical density (OD) to know the quantity of each bacterium.

Lithium Biosorption Experiment

The biosorption experiments were carried out in two phases. The first phase was to determine and demonstrate that lithium adsorption can occur in a controlled environment, and determine which bacteria species produced the most absorption under controlled conditions. The second phase was to establish the effects of the length of time (number of days) of immersion in lithium absorption.

For the first experiment, a solution with 50 ppm lithium was used to test absorption by the selected bacterial species and strains. After *E. coli* was selected as the preferred bacteria for further testing in the second phase of experiments, a lithium solution with higher concentration (150 ppm) was used to determine if absorption capacity increased with higher lithium content (>50 ppm) of the solution, and also to find out if there was any lithium toxicity and tolerance level for the selected bacteria (E. *coli*).

In the first phase of the biosorption experiments, a 50 ml lithium standard solution was prepared by adding 1.53 g of LiCl to 500 mL of distilled water to make a 500 ppm stock solution. From this 50-ppm solution, 10 ml of this stock solution was measured into a 100 ml volumetric flask and 90 ml of LB broth solution was added, 10ml was taken from the solution and 10 ml of the overnight culture contains bacteria biomass was added was added the bacterial biomass was. This was repeated for the three species/strains: Escherichia *coli*, Bacillus *subtilis* and Bacillus *cereu*s.

In the second set of experiments, a lithium solution with a concentration of 150 ppm was prepared by weighing 4.59 g of LiCl and added to 500 mL of distilled water to make a 1500 ppm stock solution of lithium, from which 10 ml was measured into a 200 ml beaker and 90 ml of cultured LB was added. Nine cups were labeled for the triplicate test experiments for the 3 inoculation time frames (24, 48 and 72 hours). The first 3 cups, labeled as (1a,1b,1c) for the first day (24 hours.), and the next 3 cups were labeled (2a 2b, 2c) for second day (48hours)while the last 3 cups for the third day were labeled (3a,3b,3c) . LB broth was produced by adding 12.5 g of LB powder into 5oo ml of distilled water. The bacteria biomass is also in the LB broth.

Ninety (90) mL of LB broth and 10mL of the lithium solution was measured into the labeled cup, from this mixture, 10 ml (10ml was taken out to obey the 9:1 serial dilution rule), then 10 ml of the overnight culture was added. This mixture was poured into all the first 3 cups labeled 1a to 1c. The same preparation was made for 2a to 2c and 3a to 3c. The pH and TDS values were measured with pH and TDS meters and the values obtained were noted. Each sample was placed in an orbital shaker at 37^{o}C 150 rpm for 24, 48, and 72 hours respectively, and the pH and TDS concentrations were checked before and after filtration with a pH/TDS meter. Samples were filtered through nanofiltration membrane (pore size 0.2 µm), and both the liquid filtrate and the biomass ascorbate were properly labeled and stored in the refrigerator. keep and properly. The liquid filtrate samples were sent out for analysis using ICP-OES in the laboratory of Dr. Gao at the University of Houston.

Lithium Recovery

The third phase experiment was for Li recovery to determine if the biomass-absorbed Li can be recovered, and the filtered biomass regenerated for re-use as absorbent. In this process, the triplicate samples of filtered biomass, for Day 2 (labeled 2a, b, c) of the previous absorbent experiment were neutralized and soaked for 12 hours in 40 ml of very dilute acid solution with a titrated pH of 4.10 to release the absorbed metals. The pH and

TDS measurements were taken before and after soaking. After the wash, the residual biomass was regenerated by adding LB broth to regrow the biomass which was later soaked in lithium solution with a known concentration. After 24 hours of re-incubation, the biomass was filtered, and the pH and TDS values of the filtrate were measured to know if lithium ions had been absorbed by the regenerated biomass.

CHAPTER 4

RESULTS AND DISCUSSION

Results of experiments carried out in this study are presented in Tables 1- 7. They represent several measurements of lithium and total dissolved solids (TDS) in lithium solutions prepared for inoculation of bacterial biomass. Total dissolved solids are often used as a measure of the concentrations of all the cations and anions n a solution and is measured in parts per million (ppm), In laboratory solutions containing lithium ion and distilled water, TDS concentrations may be a useful indication of lithium content, in the absence of direct lithium analysis. TDS was measured using calibrated TDS meters while lithium was determined by Inductively Coupled Plasma -Optical Emission Spectroscopy a (ICP-OES) at the lab of Dr Gao of the Department of Biology and Biochemistry, University of Houston, Texas.

Results were obtained with the main objectives of establishing that microbial absorption of lithium occurs in aqueous solutions, and to select which bacterial strain has the highest absorption capacity. Data was also collected to establish the effects of time on biosorption, find processes for easy recovery of adsorbed lithium and to regenerate the biomass for reuse as absorbent multiple times. Samples were prepared carefully starting with the culture and subculture of the bacterial biomass. Measurements were made carefully, and procedures were practiced multiple times before commencement of the laboratory testing. Measurements of the pH of the solutions used were routine to ensure they lie between the range of 6 and 6.5.

Reproducibility of Results

In biological and biochemical research, it is often recommended that samples be measured in triplicate to establish the reproducibility, validity, and consistency of laboratory measurements. Most of the results produced in this study were obtained in triplicate samples to ensure consistency and sample preparation and instrumental measurements, The reproducibility of the laboratory measurements from triplicate samples was estimated by using the average deviation of each measurement divided by the average value and multiplied by 100 to get the percentage deviation, The results of percent deviation for the three phases of the experiments are summarized in Tables 1 and 2.

Table 1: Percentage Variance of TDS Measurements in Triplicate Samples

	Days of Immersion	Before Filtration	After Filtration
BACILLUS subtilis	Day 1	0.11%	0.46%
	Day 2	0.23%	0.58%
	Day 3	0.46%	0.56%
BACILLUS cereus	Day 1	0.56%	0.56%
	Day 2	0.00%	0.47%
	Day 3	0.11%	5.4% *
ESCHERICHIA coli	Day 1	0.12%	0.38%
	Day 2	0.0%	0.17%
	Day 3	0.25%	0.38%

As shown in Table 1, the TDS measurements used in Phase 1 experiments in choosing the bacteria species that shows the highest absorption shows percentage variations that are mostly less than 1% except for one set of measurement of 5.6% variance that may be attributed to human error. In the Phase 2 experiments involving the choice of best absorption time there was a bit more variance in the TDS values from 2.9% to 6.7%.. The higher % variation was found in measurements after the filtration which may reflect fluctuating absorption rates by the biomass.

The lithium results show a variation of 1.6 to 8.2 % with the highest variability occurring after 3 days of inoculation of the biomass. Based on these analyses, most of the TDS and lithium measurements are within the accepted deviation rate of 5% or less. Apart from human error and instrumental instability, other factors that can account for variability in the results include biological factors in the microbial samples. The results are considered accurate, reproducible and consistent, based on procedures that were repeated numerous times.

Table 2: Variation in TDS Values to Find Best Time for Lithium Sorption

Triplicate Samples	TDS Before Filtration (ppm)	TDS (ppm) After Filtration	Lithium (ppm) ICP-OES
Day 1	2.93%	3.11%	6.92%
Day 2	5.31%	6.73%	1.62%
Day 3	4.45%	4.64%	8.28%

Evaluation of Bacterial Species

In the Phase 1 experiments, three different bacteria species were provided for evaluation (Bacillus subtilis, Bacillus cereus and Escherichia coli) to determine if they can adsorb lithium m under controlled conditions, and based on the data obtained, to select the species that shows the highest adsorption over a specified period of time. Culture biomass of these bacteria of specified quantity were inoculated in a lithium solution of known concentration over a period of 3 days during which filtrates were analyzed at 24, 48 and 72 hours intervals for TDS and Li. Results are presented in Tables 3 and 4 for TDS and Li respectively. It is apparent from the results that microbial absorption occurs in aqueous solutions, and the three bacterial types or strains absorb lithium to varying degrees.

Table 3: Total Dissolved Solids (TDS) (ppm)

TOTAL DISSOLVED SOLIDS (TDS) (ppm)										
		After 24 Hours			After 48 Hours			After 72 Hours		
		1a	1b	1c	2a	2b	2c	3a	3b	3c
Bacillus *subtilis*	Before	880	880	879	882	880	880	880	880	878
	After	877	873	875	852	856	851	853	856	858
	Difference	**-3**	**-7**	**-4**	**30**	**-24**	**-29**	**-27**	**-24**	**-20**
Bacillus *cereus*	Before	887	882	882	881	881	881	881	882	881
	After	881	880	874	851	854	855	857	810	858
	Difference	**-6**	**--2**	**-8-**	**–30**	**-27**	**-26**	**-24**	**-72**	**-23**
Escherichia *coli*	Before	801	801	800	800	800	800	802	802	802
	After	790	785	787	776	775	772	783	780	782
	Difference	**-11**	**-16**	**-13**	**-24**	**-25**	**-28**	**-19**	**-22**	**-20**

Table 4: Lithium Concentrations in Biomass Filtrate (ICP-OES)

	Lithium in Filtrate (ppm)	Lithium Absorbed (ppm)	Absorbed Li (in %)
Bacillus subtilis	22.02	27.98	55.96%
Bacillus cereus	20.44	29.56	59.12%
Escheteria coli	19.95	30.05	60.10%

The TDS results are shown in Table 3. For the first day (24 hours), the decline in TDS values was relatively low for *B. subtilis* and *B. cereus*, but higher for *E,coli* (Figure 3). However, between 24 and 48 hours (2 days), the TDS values fell considerably for all three bacterium types, but *E. coli* showed the highest total loss associated with a consistent downward trend.

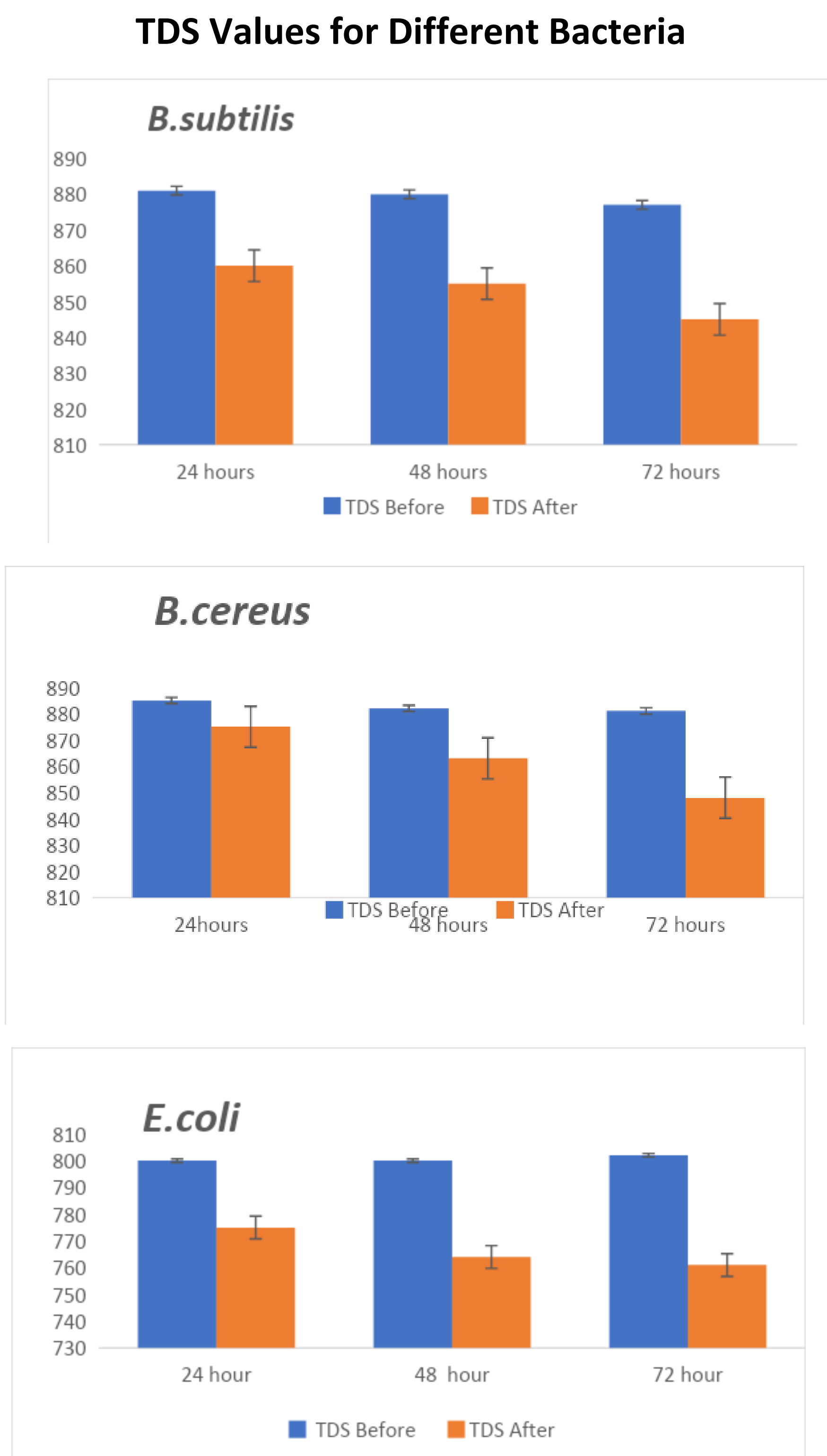

Figure 3: Charts Showing Decline in Average TDS Values for Different Bacteria

Lithium results in Table 4 suggest that E. *coli* absorbed 30 ppm Li, while B. *cereus* absorbed 29.5 ppm and *B*. *subtilis* absorbed 27.9 ppm. Using both the TDS and lithium absorption values, it is apparent that *E. coli* gave the best performance with a maximum absorption efficiency of 60% for lithium at a low concentration of 50 ppm (Table 4). Based on these experiments all the three bacteria types or strains are capable of biosorption, but E. coli was selected for further investigations in the Phase 2 experiments. It is notable that *B*. *cereus* was also very close to E. *coli* in its absorption capacity and could be used as an alternative biosorbent. This is also an indication that both gram-positive and gram-negative bacteria have the potential to absorb lithium.

Numerous studies have shown that the cell wall of microbes plays a key role in the removal of metal ions from aqueous solutions due to the presence of a great number of functional groups with different charge and geometry in the outer layers (Velkova et al., 2018) The results of this study indicate E.*coli* is a major absorber of Li in aqueous solutions which is consistent with similar behavior in the removal of cadmium (Cd) and Lead (Pb from environmental wastes. E. *coli* is a gram-negative bacterium whereas the other two are gram positive.

Most metals adsorbed by bacteria are believed to be held in the outer cell wall of the organism. Gram positive bacteria have a thick outer cell while the gram-negative bacteria have a thinner outer cell surrounded by a cell membrane which may provide a larger surface area for metal attachment (Tsurata, 2005). However, some studies (Yun et al., 2011) have suggested that some gram-positive bacteria are better absorbers and exhibited 20% more cadmium biosorption than gram-negative bacteria during bioremediation of contaminated soils. Tsuruta (2005) demonstrated that *E. coli* absorbed up to 30 ppm of Li from aqueous solutions which is comparable to results obtained in this study, it is possible that bacterial absorption in soils may be different from aqueous solutions.

With specific reference to *E. coli* as an absorbent, some studies have shown that its biomass is able to remove heavy metal ions efficiently from aquatic systems (Saleh, 2020, Sari et al., 2009). In addition, E. *coli* also has an advantage that it is ubiquitous and can be easily sophisticated and economical to use as an absorbent (Fathollahi et al., 2018; Rind et al., 2023). Therefore, E. coli is a promising and potential sorbent for Li removal from aqueous media.

Effects of Contact Time on Biosorption

Results of the Phase 2 experiments using E. *coli* biomass are used to determine the contact time in which the biomass showed maximum absorption of lithium over a period of 3 days. (24, 48 and 72 hours). Results for TDS and lithium measurements of the filtered solution are presented in Tables 5. The TDS values before and after filtration show a decline at 24, 48 and 72 hours, with the rate of declining increasing in that order (Figure 4).

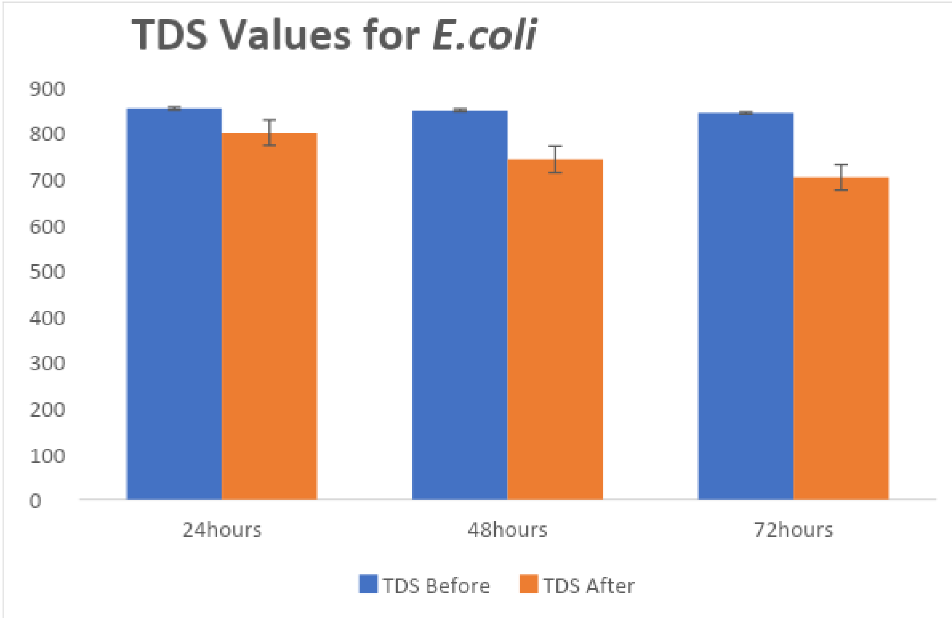

Figure 4: Chart Showing the Changes of TDS Values with Time

Table 5: TDS and Lithium Values to Evaluate Contact Time with Highest Absorption

	TDS Before Filtration (ppm)	TDS (ppm) After Filtration	Difference		**Lithium** (ppm) ICP-OES	Lithium Adsorbed	Absorption Efficiency
After 24 Hours							
Day 1a	870	814	**-56**		87.3	62.7	42%
Day 1b	845	797	**-48**		87.0	63.0	42%
Day 1c	845	789	**-56**		81.4	68.6	46%
After 48 Hours							
Day 2a	844	772	**-62**		87.0	63.0	42%
Day 2b	819	734	**-85**		86.3	63.5	42%
Day 2c	800	721	**-79**		85.6	64.4	43%
After 72 Hours							
Day 3a	802	721	**-81**		85.5	64.5	43%
Day 3b	789	700	**-80**		82.7	67.3	45%
Day 3c	767	688	**-79**		78.7	71.3	47.5

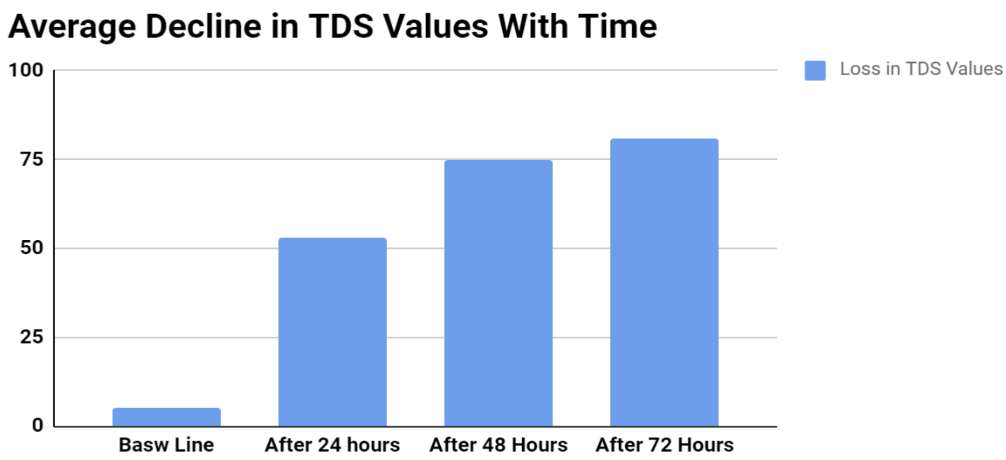

Figure 5: Chart Showing Decline in TDS Levels in Filtrate with Contact Time

TDS concentrations in the filtrate after 24 hours had decreased by values ranging from 48 to 56 ppm (mean = 52), and by 62 to 86 ppm (mean = 74) after 48 hours, and by 79 to 81 ppm (mean = 80) -after 72 hours (Figure 5). The lithium concentration in the filtrate declined fastest within the first 24 hours from 150 ppm to an average of 85 ppm after 24 hours which increased slightly to an average of 86.3 ppm after 48 hours and to 82.3 ppm after 72 hours (Figure 5). These results suggest that microbial absorption was highest after 3 days of contact time but that the rate of absorption was highest at the onset but seems to decline or stay stable after the first 24 hours. Because the experiments were limited to a maximum contract period of 3 days (72 86hours), it is not possible to establish what the maximum absorption and equilibrium were achieved.

As shown in Table 5 and Figure 6, lithium concentration declined at a high rate, from150 ppm to 87 ppm in the first 24 hours, From then on, absorption was minimal and the lithium levels fluctuate at about the same level until the 72 hours limit of the experiment ended, The amount of lithium absorbed ranged from 63 ppm to 70 ppm (Figure 7) suggesting an absorption efficiency of 43% to 47.5% (Table 5).

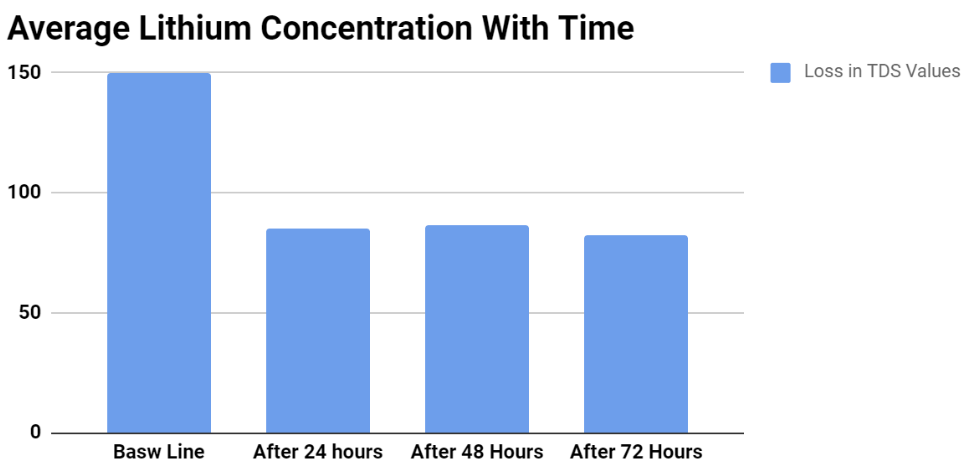

Figure 6: Decline in Lithium Levels in Filtrate with Time

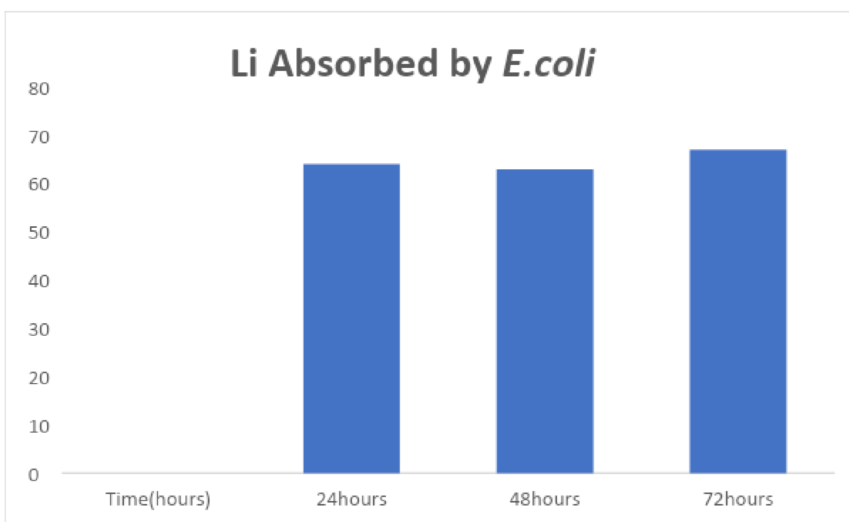

Figure 7: Lithium (in ppm) Absorbed by E. *coli* at 24, 48 and 72 Hours

Results of the majority of biosorption studies have suggested a fast initial adsorption rate of metals in aqueous solutions before the process slows and reaches an equilibrium concentration (Kalita and Joshi, 2017; Gupta and Bal Majumder, 2015) . The reason often proposed for the fast adsorption rate at the onset of the biosorption process, is the presence of more vacant and unoccupied active biosorption sites that are available on the biomass surface (Fathailahi et al., 2021). As soon as these active binding sites are occupied by metal ions the biosorption process is slowed down before reaching the saturation point and ultimately an equilibrium state (Tsai and Chen, 2010; Das et al., 2014).

Recovery of Adsorbed Lithium and Biomass Reuse Potential

One of the goals of using microbes for metal sorption is to be able to ultimately recover the metals from the absorbent biomass for beneficial end use. Phase 3 experiments were undertaken to see if a recovery of the adsorbed lithium can be achieved by using a very mild acid wash and regenerating the biomass for repeated biosorption processes. Previous test experiments undertaken in this study suggest that the maximum absorption of lithium (based on TDS measurements) occurs at a pH of 6 - 6.5. It is believed that the metals that she held can be released if the solution becomes acidic. Previous work by Tsuruta (2005) has shown that the Li metal help by biosorption was recovered by using 1M of HCl (10% HCl equivalent). The mildest acidic solution at pH 4.0 was used in the experiment. Results of a set of filtered biomass samples from Phase 2 experiments (Samples 2a, 2b and 2c) were soaked in (HNO3) acid-spiked distilled water of pH 4.10 for 12 hours to determine if lithium was released.

As shown in Table 6, the TDS values of the acid wash increased significantly from 216 ppm to above 600 ppm in triplicate samples which indicates that additional cations were released from the biomass residue (Figure 8). The biomass residue left over after the acid wash was regenerated by adding LB broth for regrowth, The regenerated biomass was inserted into a fresh lithium isolation and inoculated for 48 hours.

Table 6: pH and TDS of Acid Wash During Recovery of Lithium

Triplicate samples	Before/After Acid Wash	pH	TDS
Sample 2a	Before	4.10	216
	After	7.26	678
Sample 2b	Before	4.10	216
	After	7.23	665
Sample 2c	Before	4.10	216
	After	7.24	672
Sample 3a	Before	0.02	
	After	2.16	

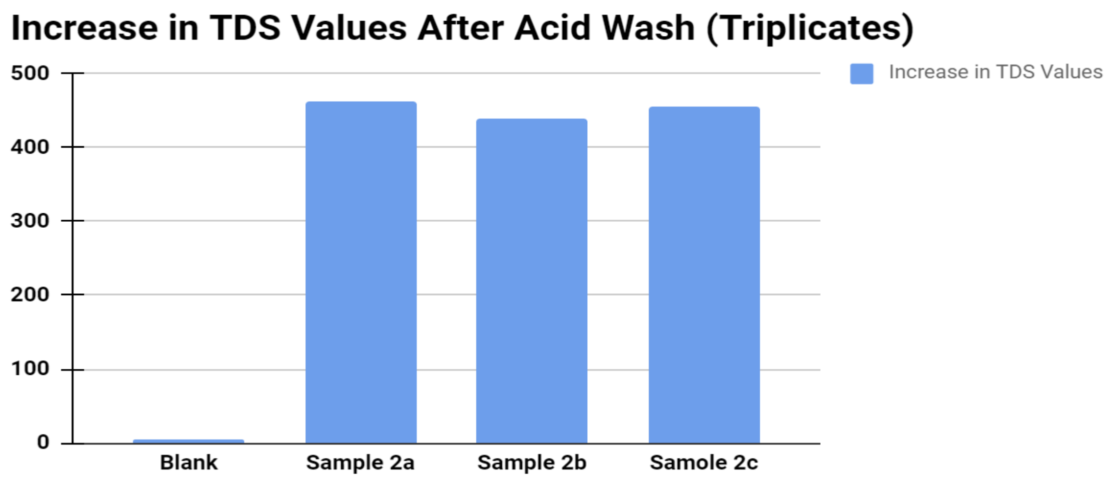

Figure 8: Chart Showing Amount of Increase in TDS Values After Acid Wash

Table 7: TDS Values Before/After Immersion of Regenerated Biomass

	Total Dissolved Solids (ppm)		
	Before Filtration	After Filtration**	pH
Sample 2a	353 358 350 —----------- Mean=353.6	365 362 333 —----------- Mean= 353.3	6.56
Sample 2b	365 362 362 —----------- Mean= 363.0	360 353 351 —----------- Mean=354.6	6.54
Sample 2c	371 368 364 —------------ Mean=367.6	353 350 349 —------------ Mean=350.6	6.56
**Regenerated biomass Inoculated in lithium solution for 48 hours			

Results of TDS measurements before and after immersion are summarized in Table 7. The pH values were also recorded to ensure they are within the range for sorption of lithium. Results indicate that there was no statistically significant increase in the TDS values after 48 hours of inoculation which suggests that lithium was not reabsorbed by the regenerated *E. coli* biomass.

There are possible explanations for these results. After the successful recovery of lithium from the filtered biomass, it is possible that it has been subjected to adverse conditions such as pH changes that could have affected its subsequent ability to reabsorb lithium during the second phase of inoculation. Microbial biosorbents are characterized by small size and low density, insufficient mechanical stability and low elasticity.to the extent that including sorbent swelling and clogging, and poor regeneration. Some of these setbacks can be remedied by immobilizing the microbial biomass, particularly for industrial applications. Immobilization will increase the physical and chemical stability of the biomass permit reus. It will physically protect the microbial biomass from environmental toxicity, enhance pH tolerance and improve physical stability and cohesion. (Velkova et al., 2018). Immobilized biomass is easily regenerated, can be used repeatedly and incorporated into fixed and fluidized bed columns which are the key elements for the practical application of biosorption (Wang et al., 2007).

CHAPTER 5

SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

This study was undertaken with several objectives including establishing that lithium, the lightest metal on Earth is subject to microbial absorption; identify from a selection of bacterial specie/strains which provides the highest absorption rate and determine the effects of contact time on lithium absorption; and attempt a recovery of absorbed lithium from biomass and explore the reuse of the regenerated biomass multiple times. Results of TDS amd lithium values from several experiments using biomass culture inoculated with lithium solutions show that *E. coli*, a gram-negative bacterium is a good absorber of lithium with an absorption capacity of up to 60% at low low lithium concentrations (<50 ppm) and up to 45% at higher lithium levels. Total dissolved solids were a reliable indicator of biosorption levels in lithium solutions. Inoculation for a period of 24 to 72 hours showed that absorption occurs rapidly within the first 24 hours, but the rate slows down considerably, although maximum absorption was achieved after 72 hours which was the time limit used in this study,

Recovery of adsorbed metals was achieved by using a very mild acid wash at a pH of 4.0 as determined by TDS measurements. The filtered residual biomass was regenerated and inoculated in a fresh lithium solution to determine if the biomass will absorb lithium. However, TDS values of the filtered solution after a 48-hour period of immersion showed there was no statistically significant difference in the results before and after filtration, which indicated that the regenerated biomass failed to resorb lithium. This failure could probably be due to exposure to changes in pH and other factors toxic to the regenerated biomass. Future experiments are recommended to involve immobilization and protection of the microbial biomass during sorption and resorption processes particularly if it involves industrial applications. Also, it is relevant to have an easily accessible and accurate method for lithium analysis, and using a lithium-ion selective electrode should be explored.

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