Notice

DESCRIPTION CN119605650A

A method for cultivating duckweed to induce massive starch accumulation

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一种诱导淀粉大量积累的浮萍培养方法

[0001]

Technical Field

技术领域

[n0001]

This invention relates to the field of aquatic energy plant cultivation, and in particular to a method for cultivating duckweed that induces a large accumulation of starch.

本发明涉及水生能源植物培养领域，特别是涉及一种诱导淀粉大量积累的浮萍培养方法。

[0003]

Background Technology

背景技术

[n0002]

Duckweed is a higher aquatic plant that grows on calm water surfaces. It mainly reproduces asexually, grows rapidly, has low requirements for water nutrients, and can absorb heavy metal ions.

浮萍是一种生长在平静水面的高等水生植物，以无性繁殖方式为主，生长速度快，对水体营养要求低，可以吸收重金属离子。

Duckweed is a rather special species of duckweed. When its growing environment lacks nutrients or the temperature is too high, it produces a starch-rich vegetative reproductive body, namely a dormant body.

多根紫萍属于浮萍的一种较为特别的物种，在生长环境缺乏营养或温度过高是，它会产生一种富含淀粉的营养繁殖体，即休眠体。

The unique structure of duckweed allows only organic matter such as starch and protein for its own metabolism to accumulate in its thallus, while synthesizing less lignin and cellulose. It has great application potential for starch extraction or fermentation to produce energy substances.

浮萍特殊的结构使得叶状体中只能积累用于自身代谢的淀粉、蛋白质等有机物，合成较少的木质素、纤维素，对于淀粉提取或发酵生产能源物质都有较大应用前景。

In addition, duckweed is rich in flavonoids such as apigenin and luteolin. In the field of traditional Chinese medicine, some duckweed (dried whole plant of duckweed or green duckweed) is also used as a medicinal material. In addition, due to the genetic stability of duckweed, current research is increasingly focusing on using duckweed as a bioreactor for developing animal vaccines.

另外，浮萍富含黄酮类物质如芹菜素、木犀草素，在我国中医药领域，也有部分浮萍(紫萍或绿萍的干燥全草)被用作中药药材。 除此之外，由于浮萍遗传稳定，现如今的研究方向逐渐将浮萍作为一种生物反应器用以开发动物疫苗。

[n0003]

Starch plays a significant role in people's daily lives and production as a primary energy source, and the required quantity is substantial.

淀粉作为主要能源物质在人们日常生产生活方面起到很大作用，所需要的量很大。

Traditional starch sources are mainly grain crops. In comparison, duckweed starch has the main advantages of faster reproduction and higher production efficiency; higher nitrogen and phosphorus utilization rate and greater environmental friendliness; unrestricted cultivation space, good adaptability, and can be cultivated in multiple layers. Although duckweed reproduces quickly and has a high starch content, its small biomass results in a low total starch production, limiting the application of duckweed starch and the development of new starch resources. Currently, there are still some problems in the research on duckweed starch. Therefore, it is necessary to provide a duckweed cultivation method that induces a large accumulation of starch to increase the biomass and starch yield of duckweed, and provide a wider range of raw material options for new starch resources.

传统淀粉来源主要是粮食作物，与之相比，浮萍淀粉的主要优势在繁殖速度快，生产效率更高；氮磷利用率高，对环境更友好；培养空间不受限，适应性好，可以多层培养。 而尽管浮萍的繁殖速度快、淀粉含量高，但是生物质量小，因此淀粉生产的总产量较低，限制了浮萍淀粉的应用，也限制了新淀粉资源的开发，目前来看对浮萍淀粉的研究尚存在一些问题，所以需要提供一种诱导淀粉大量积累的浮萍培养方法，以提高浮萍的生物量和浮萍淀粉产量，为淀粉新资源提供更广阔的原料选择。

[n0004]

Because duckweed grows quickly and requires a large space to cultivate, it is more suitable for outdoor cultivation.

浮萍由于生长速度快，培养所需空间较大，更适合在户外培养。

However, there are still some problems in the application of outdoor duckweed cultivation, including cultivation methods, industrial scale, and teaching costs. The initial investment for such equipment is relatively large, for example, it requires the installation of water systems, nutrient supply systems and other equipment. The personnel involved in the cultivation need to master the techniques of duckweed cultivation, including water quality management, nutrient supply, and pest and disease control, which is quite challenging. The lack of unified production standards and technical specifications in terms of industrial scale has led to inconsistent quality of duckweed products among different producers. This also leads to poor economic benefits for duckweed. With high investment and operating costs, the economic

return rate is not high, making it difficult for many enterprises or individuals to bear the risks of large-scale development. Meanwhile, the yield of dry matter (such as starch) in duckweed is affected by many factors, and the starch content of duckweed varies greatly, which hinders in-depth research on duckweed starch.

而在户外培养浮萍的应用过程中，培养方法、产业规模化及教学成本方面尚存在一些问题。 对于的设备初期投资较多，例如需要安装水体系统、养分供应系统等设备。 培养人员需要掌握浮萍培养的技术，包括水质管理、营养供给和病虫害防治等，难度较大。 产业规模化方面缺乏统一的生产标准和技术规范，导致不同生产者之间的浮萍产品质量参差不齐。 这也导致了浮萍的经济效益较差，在投资和运营成本较高的情况下，经济回报率不高，使得很多企业或个人难以承担规模化发展的风险。 同时浮萍也存在干物质(如淀粉)的产量受到影响因素较多、浮萍淀粉含量差距较大等原因，使得浮萍淀粉的深入研究受到阻碍。

[0007]

Summary of the Invention

发明内容

[n0005]

To address the aforementioned problems in existing technologies, this invention provides a method for cultivating duckweed that induces the accumulation of large amounts of starch.

针对现有技术存在的上述问题，本发明提供了一种诱导淀粉大量积累的浮萍培养方法。

The method for cultivating duckweed in this invention can effectively expand the cultivation of duckweed in an open outdoor environment and induce duckweed cultivation in oligotrophic environments to ultimately harvest duckweed rich in starch. This provides a new method for the research of duckweed starch resources and the large-scale cultivation practice of duckweed. It can help to better solve the problems encountered in the process of cultivating duckweed outdoors in terms of teaching costs, methods and industrial scale, and produce more starch-rich duckweed more efficiently.

本发明培养浮萍的方法能够有效通过户外开放式扩大培养浮萍、寡营养诱导浮萍最终收获富含淀粉的浮萍，为浮萍淀粉资源的研究及浮萍大规模的培养实践提供一种新方法，可以帮助更好地解决在户外培养浮萍过程中遇到的教学成本、方法和产业规模化方面的问题，更高效生产富含淀粉的浮萍。

Specifically, the present invention proposes the following technical solution:

具体来说，本发明提出了如下技术方案：

[n0006]

A method for cultivating duckweed that induces a large accumulation of starch is as follows:

一种诱导淀粉大量积累的浮萍培养方法如下：

[n0007]

Step 1, germplasm preservation: Inoculate duckweed into a solid culture medium consisting of 4.0-5.0 g/L MS solid culture medium powder, 8-12 g/L sucrose, 8-12 g/L agar, and the remainder being deionized water. The photon flux density is 80-200 μmol/m²/s, and the culture is continued.

步骤1，种质保存：将浮萍接种于固体培养基中，固体培养基组成为：4.0-5.0g/LMS固体培养基粉末、8-12g/L蔗糖、8-12g/L琼脂，其余为去离子水，光量子通量密度为80-200μmol/m²/s，持续培养；

[n0008]

Step 2, Aseptic culture: The duckweed obtained in Step 1 is inoculated into a sterile culture nutrient water. The sterile culture nutrient water is Hoagland sterile culture medium containing 10-20 g/L sucrose, and the remainder is deionized water. The pH of the water is 5.0-

5.5, and the photonic flux density is 80-200 μmol/m²/s. The culture is continued for 2-7 days.

步骤2，无菌培养：将步骤1中得到的浮萍接种于无菌培养营养水体中，无菌培养营养水体为含10-20g/L蔗糖的Hoagland无菌培养液，其余为去离子水，水体pH为5.0-5.5，光量子通量密度为80-200μmol/m²/s，持续培养2-7d；

[n0009]

Step 3, Expanded Cultivation: The duckweed obtained in Step 3 is inoculated into an expanded cultivation nutrient water body. The indoor expanded cultivation nutrient water body is a Hoagland solution diluted 4-6 times, with a water pH of 5.0-5.5; or the outdoor expanded cultivation nutrient water body is a compound fertilizer nutrient solution with a pH of 5-6, and the cultivation is continued for 3-5 days; the composition of the compound fertilizer nutrient solution is 19-23 mg/L potassium dihydrogen phosphate and 6-10 mg/L urea.

步骤3，扩大培养：将步骤3培养所得浮萍接种在扩大培养营养水体中，室内扩大培养营养水体为稀释4-6倍Hoagland溶液，水体pH为5.0-5.5；或者户外扩大培养营养水体为复配肥料营养液，pH为5-6，持续培养3-5d；所述复配肥料营养液的组成为磷酸二氢钾19-23mg/L、尿素6-10mg/L；

[n0010]

Step 4, Induction stage: Cultivate the duckweed obtained in step 3 in deionized water or tap water for 1-8 days, and harvest the duckweed thallus.

步骤4，诱导阶段：将步骤3培养所得浮萍在去离子水或自来水中，持续培养1-8d，采收浮萍叶状体。

[n0011]

The indoor cultivation process involves creating an environment most suitable for duckweed growth and starch accumulation by controlling factors such as temperature, humidity, light, nutrients, and gases under indoor conditions.

所述室内培养过程为，在室内条件下，通过控制温度、湿度、光照、营养和气体等因素，创造最适合浮萍生长和淀粉积累的环境进行浮萍培养。

The outdoor cultivation process involves using sunlight, climate conditions, natural water sources, and readily available nutrients in the outdoor natural environment to cultivate duckweed; at the same time, attention should be paid to selecting locations or water bodies with sufficient sunlight and relatively open areas.

所述户外培养过程为，利用户外自然环境中的日光、气候条件、自然水源及易获得的营养进行浮萍培养；同时注意选择日照充足、较为开阔的地点或水域。

[n0012]

In one implementation, to reduce competitive growth of algae, the surface coverage of duckweed needs to reach 45-60% during the expansion culture and induction stages.

在一种实施方式中，为减少藻类竞争生长，浮萍在扩大培养及诱导阶段的水面覆盖率需要达到45-60%。

[n0013]

In one implementation, the indoor ambient temperature of duckweed during the aseptic culture, expansion culture and induction stages is controlled at 20-30°C.

在一种实施方式中，浮萍在无菌培养、扩大培养及诱导阶段的室内环境温度控制在20-30°C。

[n0014]

In one implementation, the outdoor cultivation is irradiated by sunlight, while the indoor cultivation is irradiated by LED lights.

在一种实施方式中，户外培养的光照条件为日光，室内培养的光照为LED灯光。

[n0015]

In one implementation, outdoor-cultivated duckweed is supplemented with LED lights to provide additional illumination for 12-24 hours.

在一种实施方式中，户外培养的浮萍使用LED灯进行额外补光，满足光照时长为12-24h。

[n0016]

In one embodiment, the Hoagland nutrient solution comprises: calcium nitrate tetrahydrate 1.0-1.3 g/L, nitric acid 1.4-1.6 g/L, potassium dihydrogen phosphate 0.12-0.15 g/L, tartaric acid 2-4 mg/L, ferric chloride hexahydrate 4-7 mg/L, ethylenediaminetetraacetic acid 8-10 mg/L, magnesium sulfate heptahydrate 480-520 mg/L, sodium molybdate dihydrate 0.18-0.25 mg/L, zinc sulfate heptahydrate 0.10-0.15 mg/L, copper sulfate pentahydrate 0.05-0.12 mg/L, manganese sulfate tetrahydrate 3.30-4.00 mg/L, and a pH of 4.5-6.0.

在一种实施方式中，所述的Hoagland营养溶液组成为四水合硝酸钙1.0-1.3g/L，硝酸1.4-1.6g/L，磷酸二氢钾0.12-0.15g/L，酒石酸2-4mg/L，六水合氯化铁4-7mg/L，乙二胺四乙酸8-10mg/L，七水合硫酸镁480-520mg/L，二水合钼酸钠0.18-0.25mg/L，七水合硫酸锌0.10-0.15mg/L，五水合硫酸铜0.05-0.12mg/L，四水合硫酸锰3.30-4.00mg/L，pH为4.5-6.0。

[n0017]

Beneficial effects

有益效果

[n0018]

Compared with the prior art, the beneficial effects of the present invention are as follows:

相对于现有技术，本发明的有益效果在于：

[n0019]

1.

1.

This invention enables duckweed to accumulate starch during the induction culture process. Using these culture conditions, starch-rich duckweed thallus can be harvested quickly and efficiently.

本发明实现了浮萍在诱导培养过程中积累淀粉，利用该培养条件，可以在快速高效的收获富含淀粉的浮萍叶状体。

[n0020]

2.

2.

The method of the present invention is simple, has low equipment cost, is easy to operate, and is easy to industrialize.

本发明的方法工艺简单，设备成本低，易操作，易于实现工业化。

[n0021]

3.

3.

The method of this invention enables small, medium, and large-scale indoor or outdoor cultivation of duckweed, laying the foundation for further research and application of duckweed and expanding its development prospects.

本发明方法实现了浮萍小、中、大规模的室内或室外培养，为浮萍的进一步研究和未应用奠定了基础，拓展了浮萍的发展前景。

[0025]

Attached Figure Description

附图说明

[n0022]

Figure 1 shows duckweed being cultivated on a large scale in an outdoor plant factory in Chengdu, Sichuan Province.

图1是在四川省成都市户外植物工厂大规模培养的浮萍。

[n0023]

Figure 2 shows duckweed cultivated on a small scale outdoors in Jiaxing City, Zhejiang Province.

图2是在浙江省嘉兴市户外小规模培养的浮萍。

[n0024]

Figure 3 shows duckweed cultivated on a large scale in an indoor environment.

图3是在室内环境下大规模培养的浮萍。

[n0025]

Figure 4 shows duckweed cultivated on a small scale indoors.

图4是在室内小规模培养的浮萍。

[n0026]

Figure 5 compares the starch content of duckweed at different cultivation stages.

图5是浮萍不同培养阶段淀粉含量的比较。

[n0027]

Figure 6 shows duckweed after being dyed with starch.

图6是经过淀粉染色后的浮萍。

[n0028]

Figure 7 shows the increase in duckweed surface coverage before and after induction.

图7诱导前后浮萍水面覆盖率增长情况。

[n0029]

Figure 8 shows the increase in fresh weight of duckweed before and after induction.

图8诱导前后浮萍鲜重增长情况。

[n0030]

Figure 9 Comparison of duckweed starch yield in expanded culture and induced culture.

图9扩大培养及诱导培养的浮萍淀粉产量对比。

[n0031]

Figure 10 shows a comparison of the results of expanding duckweed cultivation outdoors using 1/5 Hoagland nutrient solution and compound fertilizer nutrient solution.

图10户外使用1/5Hoagland营养液和复配肥料营养液扩大培养浮萍的结果对比图。

[0036]

Detailed Implementation

具体实施方式

[n0032]

The preferred embodiments of the present invention are described below. It should be understood that the embodiments are for better explanation of the present invention and are not intended to limit the present invention.

以下对本发明的优选实施例进行说明，应当理解实施例是为了更好地解释本发明，不用于限制本发明。

[n0033]

Test method:

测试方法：

[n0034]

1. Method for measuring duckweed surface coverage: Take a picture of the duckweed in the entire container from the same height directly above the water surface, convert the image to 8-bit using ImageJ software, adjust the contrast appropriately, then select the duckweed leaves on the water surface using the Threshold function, and then use the Measure function to calculate the duckweed surface coverage.

1、浮萍水面覆盖率测量方法：用在相同的高度从水面正上方拍摄整个容器中的浮萍，将图片通过ImageJ软件转化类型为8-bit，适当调节对比度，然后通过Threshold功能选中水体表面的浮萍叶片，再使用Measure功能计算浮萍的水面覆盖率。

[n0035]

2. Methods for measuring duckweed quality and calculating dry matter yield: Pick out the duckweed from the water, wipe the surface and roots with kitchen paper towels to remove

moisture, and measure the fresh weight of the duckweed; after drying the harvested duckweed leaflets in an oven at 20-55°C for 8-16 hours, take them out, and measure the dry weight of the duckweed after cooling.

2、浮萍质量测量和干物质产量计算方法：将浮萍从水体中挑取出，用厨房纸巾擦拭表面及根部水分，测量浮萍鲜重；将收获的浮萍叶状体经20-55°C烘箱烘干8-16h后取出，冷却后可测量浮萍干重。

The formula for calculating duckweed yield is as follows:

浮萍产量计算公式如下：

[n0036]

Duckweed dry matter yield = dry weight ÷ (culture container area × coverage)

浮萍干物质产量＝干重÷(培养容器面积×覆盖率)

[n0037]

3. Methods for measuring the starch content of duckweed and calculating the theoretical starch yield: The dried duckweed foliage was crushed using a pulverizer and passed through a

0.2mm sieve to obtain whole duckweed foliage powder. The total starch content was determined using the Megazyme Total Starch Detection Kit. The determination method is as described in the instruction manual.

3、浮萍淀粉含量测量和淀粉理论产量的计算方法：将干燥的浮萍叶状体用粉碎机打碎，过0.2mm筛网得到浮萍叶状体全粉，用Megazyme淀粉总量检测试剂盒测定总淀粉的含量，测定方法参照说明书。

Based on the duckweed yield and starch content, the theoretical yield of duckweed starch is calculated using the following formula:

根据浮萍产量和淀粉含量得到浮萍淀粉理论产量计算公式如下：

[n0038]

Theoretical starch yield = Duckweed dry matter yield × Starch content ÷ Culture time

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淀粉理论产量＝浮萍干物质产量×淀粉含量÷培养时间

[n0039]

4. Duckweed starch staining and observation method: Duckweed was randomly sampled at different time points, soaked in 80% (v/v) ethanol solution, heated in a water bath at 40°C and shaken for 40-60 minutes to remove pigment, and washed twice with deionized water.

4、浮萍淀粉染色和观察方法：在不同时间点对浮萍进行随机取样，用80%(v/v)的乙醇溶液浸泡，水浴加热40°C震荡40-60min脱去色素，用去离子水洗涤两次。

Then stain the duckweed leaves in a 5% (v/v) Lugol solution for 2 minutes, and rinse with deionized water to remove excess staining solution.

再将浮萍叶片在5%(v/v)的Lugol溶液中染色2min，用去离子水清洗褪去多余染色液。

When observed under light, the blue area represents the region containing starch.

在光下观察，蓝色区域即为含有淀粉区域。

[n0040]

Example 1

实施例1

[n0041]

A method for cultivating duckweed to induce massive starch accumulation includes the following steps:

一种诱导淀粉大量积累的浮萍培养方法，包括如下步骤：

[n0042]

(1) Germplasm selection: The duckweed germplasm used in this study is Duckweed ZH0196, which is from the duckweed germplasm resource bank of Chengdu Institute of Biology, Chinese Academy of Sciences.

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(1)种质选择：本研究采用的浮萍种质为多根紫萍ZH0196，来源于中国科学院成都生物研究所浮萍种质资源库。

Duckweed was inoculated into a solid culture medium consisting of 4.0-5.0 g/L MS solid culture medium powder, 8-12 g/L sucrose, 8-12 g/L agar, and the remainder being deionized water. The photon flux density was 80-200 μmol/m^2/s, and the culture was continued.

将浮萍接种于固体培养基中，固体培养基组成为：4.0-5.0g/LMS固体培养基粉末、8-12g/L蔗糖、8-12g/L琼脂，其余为去离子水，光量子通量密度为80-200μmol/m2/s，持续培养。

[n0043]

(2) Aseptic culture stage: Several duckweed plants were picked from the germplasm preservation culture medium and inoculated into aseptic culture nutrient water. The aseptic culture nutrient water was Hoagland aseptic culture medium containing 10-20 g/L sucrose, and the rest was deionized water. The pH of the water was 5.0-5.5, and the photon flux density was 80-200 μmol/m²/s. The culture was continued for 2-7 days.

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(2)无菌培养阶段：将浮萍从种质保存的培养基中挑取数棵接种于无菌培养营养水体中，无菌培养营养水体为含10-20g/L蔗糖的Hoagland无菌培养液，其余为去离子水，水体pH为5.0-5.5，光量子通量密度为80-200μmol/m²/s，持续培养2-7d。

[n0044]

(3) Expanded culture of duckweed: After the duckweed in the germplasm preservation stage was activated by aseptic culture for 7 days, it was placed in 1/5 of Hoagland culture medium for open expansion culture. The culture medium composition was: calcium nitrate tetrahydrate 0.236 g/L, potassium nitrate 0.304 g/L, potassium dihydrogen phosphate 0.0272 g/L, tartaric acid 0.6 mg/L, ferric chloride hexahydrate 1.08 mg/L, ethylenediaminetetraacetic acid 1.8 mg/L, magnesium sulfate heptahydrate 0.1 g/L, sodium molybdate dihydrate 0.024 mg/L, zinc sulfate heptahydrate 0.024 mg/L, copper sulfate pentahydrate 0.016 mg/L,

manganese sulfate tetrahydrate 0.724 mg/L; the remainder was deionized water, and the pH of the water was adjusted to 5.0-5.5 with KOH or HCl.

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(3)扩大培养浮萍：将种质保存阶段的浮萍经过无菌培养阶段活化培养7d后，再放入1/5倍的Hoagland培养液中进行开放式扩大培养，培养液组成为四水合硝酸钙0.236g/L，硝酸钾0.304g/L，磷酸二氢钾0.0272g/L，酒石酸0.6mg/L，六水合氯化铁1.08mg/L，乙二胺四乙酸1.8mg/L，七水合硫酸镁0.1g/L，二水合钼酸钠0.024mg/L，七水合硫酸锌0.024mg/L，五水合硫酸铜0.016mg/L，四水合硫酸锰0.724mg/L；其余为去离子水，用KOH或HCl调节水体pH为5.0-5.5。

During the initial stage of expansion cultivation, ensure that duckweed has at least 50% water surface coverage.

扩培初始阶段保证浮萍至少50%的水面覆盖率。

The culture environment was 25°C, with a photon flux density of 80-200 μmol/m²/s, under full light for 24 hours, and cultured continuously for 3-5 days.

培养环境温度为25°C，光量子通量密度为80-200μmol/m²/s，24h全光照，持续培养3-5d。

[n0045]

(4) Indoor induction of duckweed starch accumulation: Remove the duckweed from the nutrient solution, wipe off the residual moisture on the surface, and transfer it to a culture container filled with deionized water. In the initial stage, ensure that the duckweed coverage on the water surface is about 50% and the fresh weight of the duckweed is about 2.00g.

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(4)室内诱导浮萍淀粉积累：将浮萍从营养液中取出，擦干表面残留水分，转移至盛有去离子水的培养容器中，初始阶段保证浮萍在水面的覆盖率浮萍在50%左右，浮萍的鲜重在2.00g左右。

The induction environment was set at 25°C, with a photon flux density of 80-200 μmol /m²/s, under full light for 24 hours, and induced for 2 days.

诱导环境温度为25°C，光量子通量密度为80-200μmol/m²/s，24h全光照，诱导培养2d。

[n0046]

(5) Harvesting duckweed: Before harvesting, take photos to calculate the coverage rate of duckweed; take the duckweed out of the water, wipe off the residual moisture on the surface, and weigh the fresh weight as soon as possible; then dry it in an oven at 40°C for 12 hours, and weigh it to constant weight to obtain the dry weight.

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(5)收获浮萍：收获前拍照计算浮萍的覆盖率；将浮萍从水中取出，擦干表面残留水分，尽快称量鲜重；再经过烘箱40°C烘干12h，称量至恒重得到干重。

[n0047]

Example 2

实施例2

[n0048]

(1) Expanded culture of duckweed: After the duckweed in the germplasm preservation stage was activated by sterile culture for 7 days, it was placed in 1/5 of Hoagland culture medium for open expansion culture. The composition of the culture medium was the same as in Example 1.

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(1)扩大培养浮萍：将种质保存阶段的浮萍经过无菌培养阶段活化培养7d后，再放入1/5倍的Hoagland培养液中进行开放式扩大培养，培养液组成和实例1相同。

During the initial stage of expansion cultivation, ensure that duckweed has at least 50% water surface coverage.

扩培初始阶段保证浮萍至少50%的水面覆盖率。

The culture environment was 25°C, with a photon flux density of 80-200 μmol/m²/s, under full light for 24 hours, and continuously cultured for 3-5 days.

培养环境温度为25°C，光量子通量密度为80-200μmol/m²/s，24h全光照，持续培养3-5d。

[n0049]

(2) Indoor induction of duckweed starch accumulation: Remove the duckweed from the nutrient solution, wipe off the residual moisture on the surface, and transfer it to a culture container filled with deionized water. In the initial stage, ensure that the duckweed coverage on the water surface is about 50% and the fresh weight of the duckweed is about 2.00g.

---

(2)室内诱导浮萍淀粉积累：将浮萍从营养液中取出，擦干表面残留水分，转移至盛有去离子水的培养容器中，初始阶段保证浮萍在水面的覆盖率浮萍在50%左右，浮萍的鲜重在2.00g左右。

The induction environment was set at 25°C, with a photon flux density of 80-200 μmol /m²/s, under full illumination for 24 hours, and an induction time of 20 days.

诱导环境温度为25℃，光量子通量密度为80-200μmol/m²/s，24h全光照，诱导时间为20d。

[n0050]

(3) Harvesting duckweed: Before harvesting, take photos to calculate the duckweed coverage rate; remove the duckweed leaf-like bodies from the water surface, wipe off the residual moisture on the surface, and weigh the fresh weight as soon as possible; then dry them in an oven at 40°C for 12 hours, and weigh them to constant weight to obtain the dry weight.

---

(3)收获浮萍：收获前拍照计算浮萍的覆盖率；将水面的浮萍叶状体取出，擦干表面残留水分，尽快称量鲜重；再经过烘箱40℃烘干12h，称量至恒重得到干重。

[n0051]

Example 3

实施例3

[n0052]

(1) Expanded culture of duckweed: After the duckweed in the germplasm preservation stage was activated by sterile culture for 7 days, it was placed in 1/5 of Hoagland culture medium for

open expansion culture. The composition of the culture medium was the same as in Example 1; or the duckweed was placed in compound fertilizer nutrient solution for open expansion culture. The composition of the culture medium was 21.7 mg/L potassium dihydrogen phosphate, 8.8 mg/L urea, and pH 5.5.

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(1)扩大培养浮萍：将种质保存阶段的浮萍经过无菌培养阶段活化培养7天后，再放入1/5倍的Hoagland培养液中进行开放式扩大培养，培养液组成和实例1相同；或将浮萍放入复配肥料营养液进行开放式扩大培养，培养液组成为磷酸二氢钾21.7mg/L、尿素8.8mg/L，pH为5.5。

During the initial stage of expansion cultivation, ensure that duckweed has at least 50% water surface coverage.

扩培初始阶段保证浮萍至少50%的水面覆盖率。

The culture environment was 25°C, with a photon flux density of 80-200 μmol/m²/s, under full light for 24 hours, and cultured continuously for 3-5 days.

培养环境温度为25°C，光量子通量密度为80-200μmol/m²/s，24h全光照，持续培养3-5d。

[n0053]

(2) Outdoor induction of duckweed starch accumulation: Remove the duckweed from the nutrient solution, wipe off the residual moisture on the surface, and transfer it to a culture container filled with deionized water. In the initial stage, ensure that the duckweed coverage on the water surface is about 50% and the fresh weight of the duckweed is about 2.00g.

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(2)户外诱导浮萍淀粉积累：将浮萍从营养液中取出，擦干表面残留水分，转移至盛有去离子水的培养容器中，初始阶段保证浮萍在水面的覆盖率浮萍在50%左右，浮萍的鲜重在2.00g左右。

The induction environment was outdoors, with an temperature of 16-23°C, using sunlight, and the induction time was 2 days.

诱导环境为户外，气温为16-23°C，利用太阳光照，诱导时间为2d。

[n0054]

(3) Harvesting duckweed: Before harvesting, take photos to calculate the coverage rate of duckweed; take the duckweed out of the water, wipe off the residual moisture on the surface, and weigh the fresh weight as soon as possible; then dry it in an oven at 40°C for 12 hours, and weigh it to constant weight to obtain the dry weight.

---

(3)收获浮萍：收获前拍照计算浮萍的覆盖率；将浮萍从水中取出，擦干表面残留水分，尽快称量鲜重；再经过烘箱40°C烘干12h，称量至恒重得到干重。

[n0055]

Duckweed was cultivated in different outdoor environments, as shown in Figure 1 and Figure 2: Figure 1 shows the induction of duckweed in a water tank at an outdoor plant factory in Chengdu, Sichuan Province. The water tank is 2m wide, 50m long, and 2m deep, with an isolation area in the middle of the water tank; Figure 2 shows the induction of duckweed outdoors in Jiaxing, Zhejiang Province. The cultivation container is 15cm*10cm in size, and the induction solution is 400ml.

分别在不同地方的户外环境下培养浮萍，如图1、图2所示：如图1是在四川省成都市户外植物工厂的水池中诱导浮萍，水池宽2m，长50m，深2m，在水池中间区域设有隔离；图2是在浙江省嘉兴市的户外诱导浮萍，培养容器的尺寸为15cm*10cm，诱导液均为400ml。

2

Duckweed was harvested after 2 days, and its starch content was measured to be 18.45% -32.35%.

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d后收获浮萍，测得淀粉含量为18.45%-32.35%。

[n0056]

Example 4

实施例4

[n0057]

(1) Expanded culture of duckweed: After the duckweed in the germplasm preservation stage was activated by sterile culture for 7 days, it was placed in 1/5 of Hoagland culture medium for open expansion culture. The composition of the culture medium was the same as in Example 1; or the duckweed was placed in compound fertilizer nutrient solution for open expansion culture. The composition of the culture medium was 21.7 mg/L potassium dihydrogen phosphate, 8.8 mg/L urea, and pH 5.5.

---

(1)扩大培养浮萍：将种质保存阶段的浮萍经过无菌培养阶段活化培养7d后，再放入1/5倍的Hoagland培养液中进行开放式扩大培养，培养液组成和实例1相同；或将浮萍放入复配肥料营养液进行开放式扩大培养，培养液组成为磷酸二氢钾21.7mg/L、尿素8.8mg/L，pH为5.5。

During the initial stage of expansion cultivation, ensure that duckweed has at least 50% water surface coverage.

扩培初始阶段保证浮萍至少50%的水面覆盖率。

The culture environment was 25°C, with a photon flux density of 80-200 μmol/m²/s, under full light for 24 hours, and cultured continuously for 3-5 days.

培养环境温度为25°C，光量子通量密度为80-200μmol/m²/s，24h全光照，持续培养3-5d。

[n0058]

(2) Outdoor induction of duckweed starch accumulation: The duckweed was removed from the nutrient solution and transferred to an outdoor plant factory culture pond. In the initial stage, the duckweed coverage on the water surface was at least 70%. The outdoor ambient temperature was 25-35°C. The induction time was 20 days using sunlight.

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(2)户外诱导浮萍淀粉积累：将浮萍从营养液中取出，转移至户外的植物工厂培养池中，初始阶段保证浮萍在水面的覆盖率浮萍至少为70%，户外环境温度为25-35°C，利用太阳光照，诱导时间为20d。

[n0059]

(3) Harvesting duckweed: Use a fishing net to scoop the duckweed out of the water, use a spin dryer to remove the remaining water, place it in a clean and ventilated place outdoors to dry, and then dry it in an oven at 40°C until it reaches a constant weight.

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(3)收获浮萍：使用渔网将浮萍从水面捞出，用甩干桶甩干残余水分，放在户外干净、通风处晒干，进一步经过烘箱40°C烘干至恒重。

[n0060]

The outdoor induction cultivation environment is the same as in Figure 1. Duckweed was induced in a water tank in an outdoor plant factory in Chengdu, Sichuan Province, with a water depth of 10-15 cm during the induction stage.

户外诱导的培养环境同图1，在四川省成都市户外植物工厂的水池中诱导浮萍，诱导阶段水面深度10-15cm。

Duckweed was harvested after 28 days, with a starch content of 6.32%-8.54%.

28d后收获浮萍，淀粉含量为6.32%-8.54%。

[n0061]

Example 5

实施例5

[n0062]

(1) Expanded culture of duckweed: After the duckweed in the germplasm preservation stage was activated by sterile culture for 7 days, it was placed in 1/5 of Hoagland culture medium for open expansion culture. The composition of the culture medium was the same as in Example 1. The culture environment temperature was 25°C, the photon flux density was 80-200μmol /m²/s, and the light intensity was 24h for 3-5 days.

---

(1)扩大培养浮萍：将种质保存阶段的浮萍经过无菌培养阶段活化培养7d后，再放入1/5倍的Hoagland培养液中进行开放式扩大培养，培养液组成和实例1相同；培养环境温度为25°C，光量子通量密度为80-200μmol/m²/s，24h全光照，持续培养3-5d。

[n0063]

(2) Outdoor and indoor induction of duckweed starch accumulation: Remove the duckweed from the nutrient solution, wipe off the residual moisture on the surface, and transfer it to a

culture container filled with deionized water. In the initial stage, ensure that the duckweed coverage on the water surface is about 50% and the fresh weight of the duckweed is about 2.00g.

---

(2)户外与室内诱导浮萍淀粉积累：将浮萍从营养液中取出，擦干表面残留水分，转移至盛有去离子水的培养容器中，初始阶段保证浮萍在水面的覆盖率浮萍在50%左右，浮萍的鲜重在2.00g左右。During the daytime, when the weather is sunny, the duckweed induction environment is outdoors with an air temperature of 16-23°C, utilizing sunlight; at night and on cloudy days, the induction environment is indoors with an induction temperature of 25°C, a photon flux density of 80-200μmol/m²/s, 24h full light, and an induction time of 2d.

在白天天气晴朗时浮萍诱导环境为户外，气温为16-23°C，利用太阳光照；在晚上及阴天时诱导环境为室内，诱导环境温度为25°C，光量子通量密度为80-200μmol/m²/s，24h全光照，诱导时间为2d。

[n0064]

(3) Harvesting duckweed: Use a fishing net to scoop the duckweed out of the water, use a spin dryer to remove the remaining water, place it in a clean and ventilated place outdoors to dry, and then dry it in an oven at 40°C until it reaches a constant weight.

---

(3)收获浮萍：使用渔网将浮萍从水面捞出，用甩干桶甩干残余水分，放在户外干净、通风处晒干，进一步经过烘箱40°C烘干至恒重。

[n0065]

The indoor environment for cultivating duckweed is shown in Figures 3 and 4.

室内培养浮萍环境如图3和图4所示。

Duckweed was harvested after 2 days, with a starch content of 34.60±0.47%.

2d后收获浮萍，淀粉含量为34.60±0.47%。

[n0066]

Example 6: Duckweed cultured outdoors using compound fertilizer nutrient solution

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实施例6户外使用复配肥料营养液扩大培养的浮萍

[n0067]

(1) After the duckweed in the germplasm preservation stage was activated by sterile culture for 7 days, it was then placed in a compound fertilizer nutrient solution for open-type expansion culture. The culture solution consisted of 21.7 mg/L potassium dihydrogen phosphate, 8.8 mg/L urea, and pH 5.5.

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(1)将种质保存阶段的浮萍经过无菌培养阶段活化培养7d后，再放入复配肥料营养液进行开放式扩大培养，培养液组成为磷酸二氢钾21.7mg/L、尿素8.8mg/L，pH为5.5。

In the initial stage of expansion cultivation, ensure that the duckweed has a water surface coverage of at least 50% and a fresh weight of about 2.00g.

扩培初始阶段保证浮萍至少50%的水面覆盖率，浮萍的鲜重在2.00g左右。

The propagation environment is outdoors, with a temperature of 16-23°C, utilizing sunlight in the environment.

扩培环境为户外，气温为16-23°C，利用环境中太阳光。

[n0068]

(2) After harvesting duckweed, measure the coverage rate and fresh weight yield.

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(2)收获浮萍后测量覆盖率，鲜重产量。

[n0069]

Comparative Example 1: Duckweed during the germplasm preservation stage:

对比例1种质保存阶段的浮萍：

[n0070]

Prepare solid MS medium with the following composition: 4.0-5.0 g/L MS solid medium powder, 8-12 g/L sucrose, 8-12 g/L agar, and the remainder being deionized water.

配制固体MS培养基，其组成为：4.0-5.0g/L MS固体培养基粉末、8-12g/L蔗糖、8-12g/L琼脂，其余为去离子水。

The prepared culture medium was placed in an autoclave and sterilized at 115°C for 20 minutes.

配好的培养基放入高压灭菌锅，在115℃下灭菌20min。

Once the MS medium temperature has dropped to around 50°C, pour it into plates while it is still hot, about 25 mL per plate.

待MS培养基温度降至50°C左右，趁热倒入平板，每板倒约25mL培养基。

After it cools and solidifies in the laminar flow hood, pick out healthy, sterile duckweed and place it on a flat plate, then seal it with sealing film.

待其在超净台内冷却凝固后，挑取健康的无菌浮萍到平板中，用封口膜封口。

The ambient temperature was 25°C, the light-dark cycle was 16:8, the photon flux density was 80-200 μmol/m²/s, and the culture was carried out for 14 days.

环境温度为25°C，光暗周期为16:8，光量子通量密度为80-200μmol/m²/s，持续培养14天。

The duckweed was dried at 40°C for 12 hours, ground into powder, and the starch content was measured using a reagent kit.

将浮萍经40°C烘干12h，磨粉，用试剂盒法测量淀粉含量。

[n0071]

Comparative Example 2: Duckweed during the aseptic culture stage:

对比例2无菌培养阶段的浮萍：

[n0072]

Prepare liquid Hoagland medium with the following nutrient solution composition: calcium nitrate tetrahydrate 1.18 g/L, potassium nitrate 1.52 g/L, potassium dihydrogen phosphate 0.136 g/L, tartaric acid 3 mg/L, ferric chloride hexahydrate 5.4 mg/L, ethylenediaminetetraacetic acid 9 mg/L, magnesium sulfate heptahydrate 0.5 g/L, sodium molybdate dihydrate 0.12 mg/L, zinc sulfate heptahydrate 0.12 mg/L, copper sulfate pentahydrate 0.08 mg/L, and manganese sulfate tetrahydrate 3.62 mg/L.

配制液体Hoagland培养基，营养溶液组成为四水合硝酸钙1.18g/L，硝酸钾1.52g/L，磷酸二氢钾0.136g/L，酒石酸3mg/L，六水合氯化铁5.4mg/L，乙二胺四乙酸9mg/L，七水合硫酸镁0.5g/L，二水合钼酸钠0.12mg/L，七水合硫酸锌0.12mg/L，五水合硫酸铜0.08mg/L，四水合硫酸锰3.62mg/L。

Add 1.5% (w/w) sucrose to the culture medium, with the remainder being deionized water, and adjust the pH of the water to 5.0-5.5 with KOH or HCl.

并在培养液中加入1.5%(w/w)的蔗糖，其余为去离子水，用KOH或HCl调节水体pH为5.0-5.5。

The prepared culture medium was placed in an autoclave and sterilized at 115°C for 20 minutes.

配好的培养基放入高压灭菌锅，在115℃下灭菌20min。

After the culture medium cools down, sterile duckweed germplasm is inoculated. The culture environment temperature is 25°C, the photonic flux density is 80-200μmol/m²/s, and the culture is continued for 2-7 days.

待培养液冷却后，接种无菌浮萍种质，培养环境温度为25℃，光量子通量密度为80-200μmol /m²/s，持续培养2-7d。

The duckweed was dried at 40°C for 12 hours, ground into powder, and the starch content was measured using a reagent kit.

将浮萍经40℃烘干12h，磨粉，用试剂盒法测量淀粉含量。

[n0073]

Comparative Example 3: Duckweed in the indoor extended culture stage:

对比例3室内扩大培养阶段的浮萍：

[n0074]

Prepare a nutrient culture medium consisting of 1/5 times the volume of Hoagland medium. The medium composition is as follows: calcium nitrate tetrahydrate 0.236 g/L, potassium nitrate 0.304 g/L, potassium dihydrogen phosphate 0.0272 g/L, tartaric acid 0.6 mg/L, ferric chloride hexahydrate 1.08 mg/L, ethylenediaminetetraacetic acid 1.8 mg/L, magnesium sulfate heptahydrate 0.1 g/L, sodium molybdate dihydrate 0.024 mg/L, zinc sulfate heptahydrate 0.024 mg/L, copper sulfate pentahydrate 0.016 mg/L, and manganese sulfate tetrahydrate 0.724 mg/L. The remainder is deionized water. Adjust the pH of the water to 5.0-5.5 using KOH or HCl.

配制营养培养液，组成为1/5倍的Hoagland培养液，培养液组成为四水合硝酸钙0.236g/L，硝酸钾0.304g/L，磷酸二氢钾0.0272g/L，酒石酸0.6mg/L，六水合氯化铁1.08mg/L，乙二胺四乙酸1.8mg/L，七水合硫酸镁0.1g/L，二水合钼酸钠0.024mg/L，七水合硫酸锌0.024mg/L，五水合硫酸铜0.016mg/L，四水合硫酸锰0.724mg/L；其余为去离子水，用KOH或HCl调节水体pH为5.0-5.5。

Remove the sterile cultured duckweed from the nutrient solution and conduct open culture in a container of appropriate size, maintaining at least 50% water surface coverage.

将无菌培养的浮萍从营养液中取出，在适当尺寸容器中进行开放式培养，保持至少50%的水面覆盖率。

The culture environment was 25°C, the photon flux density was 80-200μmol/m²/s, and the culture was carried out for 4 days.

培养环境温度为25°C，光量子通量密度为80-200μmol/m²/s，持续培养4d。

The duckweed was dried at 40°C for 12 hours, ground into powder, and the starch content was measured using a reagent kit.

将浮萍经40°C烘干12h，磨粉，用试剂盒法测量淀粉含量。

[n0075]

Comparative Example 4: Duckweed cultured outdoors using 1/5 Hoagland medium:

对比例4户外使用1/5Hoagland培养液扩培的浮萍：

[n0076]

After the duckweed in the germplasm preservation stage was activated by aseptic culture for 7 days, it was then placed in 1/5 of Hoagland culture medium for open expansion culture. The composition of the culture medium was the same as that of comparative example 3.

将种质保存阶段的浮萍经过无菌培养阶段活化培养7d后，再放入1/5倍的Hoagland培养液中进行开放式扩大培养，培养液组成同对比例3。

Open cultivation should be carried out in appropriately sized containers, maintaining at least 50% water surface coverage, with the fresh weight of duckweed around 2.00g.

在适当尺寸容器中进行开放式培养，保持至少50%的水面覆盖率，浮萍的鲜重在2.00g左右。

The culture environment is outdoors, with an temperature of 16-23°C, and the culture is carried out continuously for 4 days using sunlight.

扩培环境为户外，气温为16-23°C，利用环境中太阳光，持续培养4天。

After harvesting duckweed, measure the water surface coverage and fresh weight yield.

收获浮萍后测量水面覆盖率，鲜重产量。

[n0077]

Figure 5 is a comparison chart of the starch content of duckweed in Comparative Examples 1, 2, and 3 and the Example.

图5为对比例1、2、3与实施例的浮萍淀粉含量对比图。

As shown in Figure 5, the starch content was very low during the germplasm culture, aseptic culture, and scale-up culture stages, with the starch content ranging from 9.76% to 11.11%, 4.85% to 5.52%, and 1.94% to 4.51%, respectively.

由图5可知，在种质培养、无菌培养以及扩大培养阶段淀粉含量很低，淀粉含量的范围依次为9.76%-11.11%、4.85%-5.52%以及1.94%-4.51%。

In this example, the starch content of the induced duckweed can reach 45.03%, which is 4.9-23 times that of the comparative example.

而在实施例这诱导过后的浮萍的淀粉含量可以达到45.03%，是对比例的4.9-23倍。

This indicates that the starch content of duckweed can be significantly increased after induction.

说明浮萍淀粉经过诱导后可以显著提高淀粉含量。

[n0078]

Figure 6 is a comparison of the starch staining of duckweed in Comparative Example 3 and Example 1.

图6为对比例3与实施例1的浮萍淀粉染色对比图。

As shown in Figure 6, duckweed starch accumulates in large quantities in the leaves during the early induction period.

由图6可知，诱导前期浮萍淀粉在叶片中大量积累。

This indicates that the starch content of duckweed can be significantly increased after induction.

说明浮萍淀粉经过诱导后可以显著提高淀粉含量。

[n0079]

Figure 7 shows the duckweed surface coverage before and after induction in Examples 1, 3, and 5, to determine the growth of duckweed.

图7为实施例1、实施例3、实施例5在诱导前后浮萍水面覆盖率情况，以此来判断浮萍的增长情况。

As shown in Figure 7, duckweed continued to grow under all three conditions even though nutrients were reduced within two days of induction.

由图7可知，诱导2天内，虽然营养减少，但是三种情况下浮萍仍在生长。

Moreover, the growth of duckweed cultivated outdoors and indoors is similar to that of duckweed cultivated indoors; at the same time, the growth rate of duckweed cultivated outdoors alone is slightly lower than the other two cases, but it should be considered that poor outdoor light conditions may have a certain negative impact on duckweed growth.

而且户外+室内培养浮萍生长情况与室内培养浮萍生长情况接近；同时单户外培养的浮萍生长速度略低于其他两种情况，但是需要考虑到户外光照情况较差可能会对浮萍生长产生一定负面影响。

The above demonstrates that outdoor cultivation of induced duckweed is feasible and can maintain strong growth vitality in duckweed.

以上说明户外培养诱导浮萍是可行的，能够保持浮萍较强的生长活力。

[n0080]

Figure 8 shows the changes in fresh weight of duckweed before and after induction in Examples 1, 3, and 5, which can be used to further quantify the growth of duckweed.

图8为实施例1、实施例3、实施例5在诱导前后浮萍鲜重的变化情况，以此可以进一步量化浮萍的增长情况。

As shown in Figure 8, the accumulation of duckweed material increased again within 2 days of induction.

由图8可知，诱导2天内，浮萍物质积累均再增加。

Similar to the water surface coverage situation, the growth of duckweed cultivated outdoors and indoors is close to the fresh weight of duckweed cultivated indoors; however, the growth rate of duckweed cultivated outdoors alone is lower than the other two situations.

与水面覆盖率情况类似，户外+室内培养浮萍生长情况与室内培养浮萍鲜重接近；而单户外培养的浮萍生长速度要低于其他两种情况。

This indicates that outdoor lighting combined with supplemental lighting provides a better induction effect.

说明户外+补光诱导效果更好。

[n0081]

Figure 9 shows a comparison of duckweed starch yield in expanded culture (Comparative Example 3) and induced culture (Examples 1, 3, and 5).

图9为在扩大培养(对比例3)及诱导培养(实施例1、实施例3、实施例5)的浮萍淀粉产量对比图。

This indicates that oligotrophic induction can significantly increase starch production in duckweed thallus, whether indoors or outdoors. Although the outdoor + indoor induction method is not as good as completely indoor culture, it is still better than completely outdoor culture.

说明无论是在室内还是在户外，寡营养诱导均可以显著提高浮萍叶状体淀粉产量，相比来说，尽管户外+室内的诱导形式不如完全室内培养，但是也要优于完全户外培养。

[n0082]

Figure 10 is a comparison of the results of expanding duckweed cultivation outdoors using 1/5 Hoagland nutrient solution (Comparative Example 4) and compound fertilizer nutrient solution (Example 6).

图10为在户外使用1/5Hoagland营养液(对比例4)和复配肥料营养液(实施例6)扩大培养浮萍的结果对比图。

Under outdoor conditions, the effect of using compound fertilizer nutrient solution for expansion culture for 4 days is better, with water surface coverage reaching 100% and fresh weight also higher.

在户外条件下，使用复配肥料营养液扩培4天后的效果更好，水面覆盖率达到100%，鲜重也更高。

This may be due to the significant variations in factors such as sunlight, temperature, humidity, and wind speed under outdoor conditions, which may cause the nutrients in Hoagland nutrient solution to be rapidly consumed or degraded, making it unable to continuously provide the nutrients needed by the plants.

这可能是由于户外条件下的光照、温度、湿度及风速等因素变化较大，可能导致Hoagland营养液的营养成分被快速消耗或降解，无法持续提供植物所需的营养。

Compound fertilizers may be better adapted to this dynamic environment and can release nutrients for a longer period of time.

而复配肥料可能更适应这种动态环境，能够更持久地释放营养。

In addition, in outdoor environments, 1/5 Hoagland nutrient solution is more susceptible to the influence of microorganisms and algae, which may compete with duckweed for nutrients; while compound fertilizers may inhibit the growth of these organisms to some extent, thus better supporting the growth of duckweed.

另外，户外环境中，1/5Hoagland营养液更容易受到微生物和藻类的影响，这些生物可能会与浮萍竞争营养；而复配肥料可能在一定程度上抑制这些生物的生长，从而更好地支持浮萍的生长。

[n0083]

Although the present invention has been disclosed above with reference to preferred embodiments, it is not intended to limit the present invention. Anyone skilled in the art can make various modifications and alterations without departing from the spirit and scope of the present invention. Therefore, the scope of protection of the present invention should be determined by the claims.

虽然本发明已以较佳实施例公开如上，但其并非用以限定本发明，任何熟悉此技术的人，在不脱离本发明的精神和范围内，都可做各种的改动与修饰，因此本发明的保护范围应该以权利要求书所界定的为准。