

Opinion

# A Proposal on the Restoration of *Nostoc flagelliforme* for Sustainable Improvement in the Ecology of Arid Steppes in China

Xiang Gao <sup>1,\*</sup>, Haiyan Xu <sup>1</sup>, Shuifeng Ye <sup>2</sup> and Wenyu Liang <sup>3,\*</sup>

<sup>1</sup> School of Life Sciences, Hubei Key Laboratory of Genetic Regulation and Integrative Biology, Central China Normal University, Wuhan 430079, China; designer1991xhy@126.com

<sup>2</sup> Shanghai Agrobiological Gene Center, Shanghai 201106, China; ysf@sagc.org.cn

<sup>3</sup> School of Life Sciences, Ningxia University, Yinchuan 750021, China

\* Correspondence: xianggao@mail.ccnu.edu.cn (X.G.); liang\_wy@nxu.edu.cn (W.L.);

Tel.: +86-27-6786-7223 (X.G.); +86-951-2062051 (W.L.)

Academic Editor: Yu-Pin Lin

Received: 20 April 2016; Accepted: 24 May 2016; Published: 2 June 2016

**Abstract:** *Nostoc flagelliforme*, a filamentous nitrogen-fixing cyanobacterium, is widely distributed in arid steppes of the west and northwestern parts of China. However, as a food delicacy this species has been overexploited from 1970 to 2000. Moreover, overgrazing, land reclamation and the removal of medicinal herbs have caused severely reduced vegetation coverage there. In this communication, a badly damaged but slowly rehabilitating *N. flagelliforme*-inhibiting steppe is described, and the rehabilitation of desertified steppes by the renewed growth of *N. flagelliforme* is proposed. The restoration of this dominant nitrogen supplier would be an ecologically sustainable solution for supplementing current measures already taken in the desertified regions. In addition, a goal of 50%–60% vegetation coverage is proposed by the *N. flagelliforme* restoration.

**Keywords:** terrestrial cyanobacteria; *Nostoc flagelliforme*; arid steppe; carbon-nitrogen cycle; ecological improvement

## 1. Reduced Biomass of *Nostoc flagelliforme* as an Indicator of Desertification

Terrestrial cyanobacteria are the dominant population of algae in ecologically fragile desert steppes. They contribute organic nitrogen (N) and carbon (C) resources for the nutrient-poor soils [1]. In the arid steppes of the west and northwestern parts of China, some of them, such as *Microcoleus* species, participate in the formation of biological soil crusts (BSCs), which are both widespread and important for sand fixation [2,3]. Another widespread cyanobacterium is *Nostoc flagelliforme*, which appears as a hair-like colony form on the soil surface [4]. As a popular food delicacy, *N. flagelliforme* had been excessively exploited by raking from 1970s to 2000 in China [5]. A good example is Ningxia Province, in which the producing area for *N. flagelliforme* was about  $360.7 \times 10^4$  hm<sup>2</sup> in the 1960s, but was reduced to  $173.6 \times 10^4$  hm<sup>2</sup> in the mid-1980s, while the biomass was reduced from 3 to 7.5 kg/hm<sup>2</sup> in the 1970s to 0.15 kg/hm<sup>2</sup> in the 1990s [6]. In order to rehabilitate desertified regions, some preventative measures were implemented around 2000 in China, including prohibition from grazing, returning sloping field to grassland and prohibiting the trade of *N. flagelliforme*. This has resulted in some positive effects [7,8].

In arid steppes, soil N is usually considered as being secondary to water content in limiting ecological processes. The natural ecological recovery of the N supplier *N. flagelliforme* as well as vegetation coverage should be a very long process. *N. flagelliforme* has less than a 6% annual growth rate [9]. However, we found that some local governments were more inclined to accept

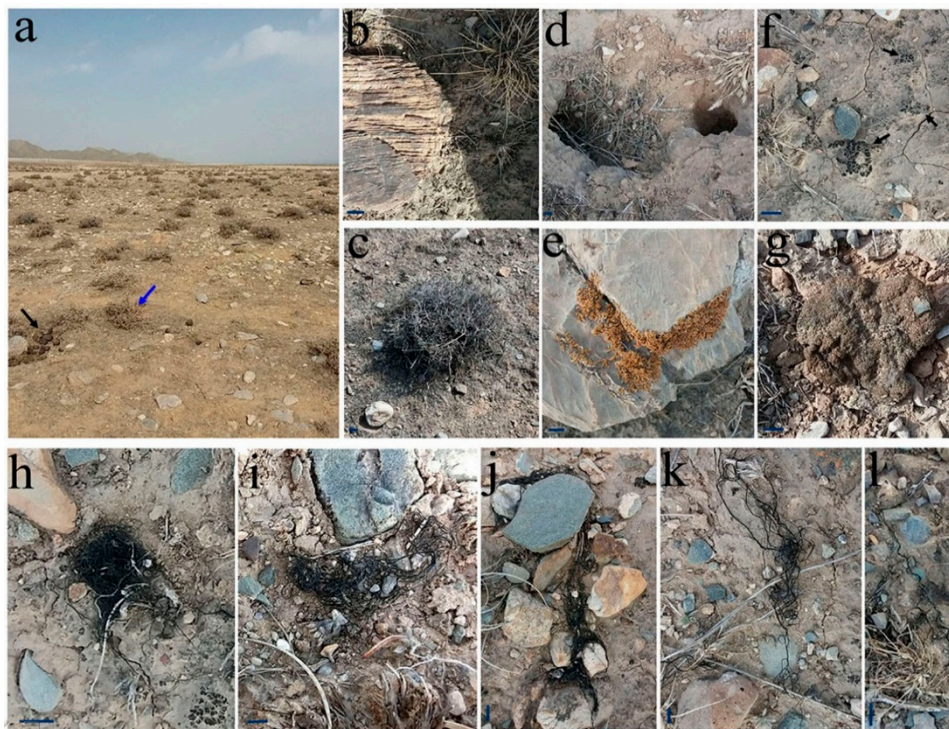
immediate measures such as grass planting or afforestation for combating desertification, rather than the restoration of N suppliers such as *N. flagelliforme*. Such a “success” may be temporary or unsustainable [10]. In addition, it was reported that exogenous N additions exhibited neutral or even inhibitory effects on plant abundance and litter decomposition [11,12]. Therefore, an understanding of the dynamic equilibrium of C and N suppliers in arid steppes is of importance for sustained rehabilitation of damaged soils. In this communication, we describe a badly damaged but slowly rehabilitating arid steppe, while also emphasizing the great importance of *N. flagelliforme* restoration for achieving combined recovery of this cyanobacterium and xerophilic plants for ecological improvement in the arid steppes.

## 2. A Badly Damaged but Slowly Rehabilitating *N. flagelliforme*-Inhabiting Steppe

A scene of this rehabilitating steppe in winter is shown in Figure 1. Dwarfish shrubs were scattered throughout the whole steppe, with less than 5% vegetation coverage (Figure 1a). The vegetation coverage in the rainy season (summer) is about 15%–30%, which indicates moderate desertification [13]. Horse feces were occasionally found, suggesting that there is grazing disturbance (Figure 1a). The soils are calcic sierozem and light chestnut with pH of 8–9.5 [4,6], as implied by the weathered stones (Figure 1b). The most abundant two plants in this steppe were *Ephedra lepidosperma* C.Y. Cheng (Figure 1c) and *Stipa breviflora* Griseb (in Figure 1b). Approximately 10–20 withered plant species on the surface soil were found. In other regions with *N. flagelliforme* distribution, plant diversity may be different [6]. Around 40 or more plant species could be found in the rainy summer season in Ningxia Province (Figure S1). These higher plants serve as the main C supplier in arid steppes, since soil C was positively correlated to vegetation coverage and above-ground biomass, especially surface litter [14]. Traces of rodent activity were found as suggested by their holes (Figure 1d). Surveys showed that *Meriones unguiculatus* and *Meriones meridianus* were the two dominant rat species [15]. Proper animal activity, including controlled grazing intensity, was conducive to the virtuous cycle of ecology [16,17].

In addition, colonizing pioneers (also N suppliers) such as lichens (Figure 1e), BSCs (Figure 1f–g), and *N. flagelliforme* colonies (Figure 1h–l) were observed. Lichens were occasionally found, adhering to bare stones (Figure 1e). *N. flagelliforme* colonies were tightly (Figure 1h) or loosely (Figure 1i) aggregated, or mostly stretched by surface runoff (Figure 1j–l), entwining around stones or grasses. Most of these *N. flagelliforme* colonies were about 10–20 cm long, far from the length of 50 cm or more that they can grow up to. On the whole, small BSCs and *N. flagelliforme* colonies were widely but sparsely spread, suggesting that many had been destroyed or collected.

*N. flagelliforme* yield had reduced by at least 95% because of the over-collection. In addition, raking *N. flagelliforme* from the soil surface would have severely affected the development of BSCs. Overgrazing, land reclamation and the removal of medicinal herbs (e.g., *Glycyrrhiza uralensis* and *Herba ephedrae*) also accounted for the severe reduction of vegetation coverage and quality [18–20]. Therefore, this desolate scene reflects the comprehensive damage to *N. flagelliforme*, BSC and vegetation on the arid steppe. It also hints that soil C–N dynamics is currently at a weak equilibrium. Instead, a combined recovery of C suppliers (shrubs and grasses) and N suppliers (*N. flagelliforme* and BSCs) may be necessary for steppe rehabilitation.



**Figure 1.** A slowly rehabilitating *N. flagelliforme*-inhabiting steppe in Yinchuan, Ningxia Province, China. (a) The steppe scene in winter. Blue arrow points to *Ephedra lepidosperma* C.Y. Cheng (shrub); black arrow points to animal feces; (b) the weathered stone. The adjacent plants are *Stipa breviflora* Griseb; (c) the dominant shrub in this steppe, *E. lepidosperma* C.Y. Cheng; (d) the rat holes; (e) the lichens in a stone; (f) small BSCs; (g) a big BSC; (h) the aggregated *N. flagelliforme* colonies; (i) the loosely aggregated *N. flagelliforme* colonies; (j–l) the stretched *N. flagelliforme* colonies by surface runoff. BSCs, biological soil crusts. Bar, 1 cm.

### 3. The Potential of the Restoration of *N. flagelliforme* for Ecology Improvement

In the dynamic soil C–N cycle in arid steppes, xerophilic plants contribute to the majority of C storage [14,21]. Soil microorganisms and enzyme activities (e.g., polyphenol oxidase, cellulase,  $\beta$ -glucosidase, nitrate reductase and urease), which play crucial roles in the cycle, varied in correlation with the vegetation type and coverage [22,23]. However, the current low N level still remains a limiting factor for plant growth. Soil N level is only 0.7 g/kg in the aforementioned steppe and the average level in Ningxia is 0.47 g/kg [6]. An interesting finding is that the biomass of *N. flagelliforme* in soils with relatively rich organic matter (e.g., 1.0%) is higher than in those with less organic matter (e.g., 0.02%) [24,25]. Laboratory cultivation found that *N. flagelliforme* has the capability to use organic carbon sources for heterotrophic growth in darkness [26,27]. Thus, the organic products decomposed by soil bacteria from plant litter may sustain the relatively superior growth of *N. flagelliforme*. The development of BSCs could also be promoted by plantation establishment [28]. Therefore, an interactive promotion in growth between C and N suppliers actually exists. In the N-poor and relatively C-rich soils, it is possible that *N. flagelliforme* shifts its two roles (C and N suppliers) to one main role (N supplier) to collaborate with its surrounding biological circumstances.

An engineering experiment in Inner Mongolia, China, has proved that the “algae–grass–shrub” strategy, using the restoration of BSC, is feasible for accelerating the reversal of desertified land [29,30]. Similarly, a dynamic combined recovery of *N. flagelliforme* resource and vegetation coverage could be imagined by the restoration of *N. flagelliforme*. According to historical data [13], summer vegetation coverage of 50%–60% is expected to be steadily achieved through *N. flagelliforme* restoration coupled with other protective measures. *N. flagelliforme* is photophilic, since photosynthesis of rewetted

*N. flagelliforme* is saturated at 1000  $\mu\text{mol photons m}^{-2}\cdot\text{s}^{-1}$  and no photoinhibition is recognized until 1800  $\mu\text{mol photons m}^{-2}\cdot\text{s}^{-1}$  [4]. Owing to this feature, the dispersed density (not biomass) of *N. flagelliforme* colonies would be affected at higher vegetation coverage. However, temperate soil algae and shrubs would appear more in those circumstances as suggested by Liu [29], thus maintaining a higher level of soil C–N cycling. An enhanced soil C–N cycle will also serve to maintain an ecological stability upon climate changes [31].

Compared to the restoration of BSC for ecological improvement, the use of *N. flagelliforme* propagules may be more convenient for dispersion and development on surface soils. Several ways have been suggested for reproduction of *N. flagelliforme*: (1) single cells or small filaments fragmented from big filaments forming new colonies; (2) via akinetes; (3) hormogonia dispersion and formation of new colonies [4]. The fragmentation of filaments to a very small size is an easy way to prepare propagules for direct application in natural habitats, but this will consume a huge amount of natural colonies.

Alternatively, mass cultivation of *N. flagelliforme* under aquatic conditions is an efficient way of preparing a large number of propagules [32,33]. However, a significant problem for these liquid-cultured propagules is that they are not very resistant to environmental stresses, such as desiccation stress, and may need extra processing, such as wrapping them with stabilizing components [33]. The solution of this key problem will greatly accelerate the application of *N. flagelliforme* for the improvement of ecology in arid steppes. According to the 6% annual growth rate, the growth of the filaments from 1 cm to the expected 50–60 cm in length still needs about 70 years. Therefore, the restoration of *N. flagelliforme* and the whole ecosystem is a long-term process.

#### 4. Conclusions

This communication has proposed the restoration of *N. flagelliforme* for ecological improvement of arid steppes. This restoration may accelerate the recovery of vegetation cover and its quality, while it should be more ecologically sustainable compared to only planting xerophilic plants. In the current desertification situation, this restoration may also provide a valuable supplement for the measures already taken in desertification rehabilitation. In addition, the existing problem is that we have not completely mastered the biotechnology for cultivating *N. flagelliforme* propagules that are resistant to environmental stresses. Once this problem is overcome, it would be important to run field experiments or even to attempt large-scale application of *N. flagelliforme* propagules.

**Supplementary Materials:** The following are available online at [www.mdpi.com/2076-3298/3/2/14/s1](http://www.mdpi.com/2076-3298/3/2/14/s1), Figure S1. Shrubs and grasses in rainy season in the eastern side of the Helan Mountain in Yinchuan, Ningxia Province, P. R. China. (1) *Sarcozygium xanthoxylon* Bunge; (2) *Zygophyllum mucronatum* Maxim.; (3) *Dracocephalum heterophyllum* Benth.; (4) *Artemisia sacrorum* Ledeb.; (5) *Aristida adscensionis* L.; (6) *Euphorbia humifusa* Willd. ex Schlecht.; (7) *Thymus mongolicus* Ronn.; (8) *Convolvulus tragacanthoides* Turcz.; (9) *Ephedra Lepidosperma* C.Y. Cheng; (10) *Halogoton arachnoideus* Moq.; (11) *Tragus racemosus* (L.) All.; (12) *Cynanchum thesioides* (Frey) K. Schum.; (13) *Cleistogenes squarrosa* (Trin.) Keng; (14) *Anabasis brevifolia* C.A. Mey.; (15) *Lagochilus ilicifolius* Bunge; (16) *Reaumuria trigyna* Maxim.; (17) *Gentiana dahurica* Fischer; (18) *Reaumuria soongorica* (Pall.) Maxim.; (19) *Sibbaldia adpressa* Bge.; (20) *Asparagus gobicus* Ivan. ex Grubov; (21) *Enneapogon borealis* (Griseb.) Honda; (22) *Potentilla acaulis* L.; (23) *Ajania fruticulosa* (Ledeb.) Poljak.; (24) *Chloris virgata* Sw.; (25) *Peganum harmala* L.; (26) *Peganum nigellastrum* Bge.; (27) *Caryopteris mongholica* Bge.; (28) *Eragrostis minor* Host; (29) *Convolvulus ammannii* Desr.; (30) *Plantago minuta* Pall.; (31) *Gypsophila davurica* Turcz. ex Fenzl.; (32) *Linum perenne* L.; (33) *Salsola laricifolia* Turcz. ex Litv.; (34) *Ixeris denticulate*; (35) *Iris tenuifolia* Pall.; (36) *Stellariae dichotoma* L. var. *lanceolata* Bge.; (37) *Scorzonera divaricata* Turcz.; (38) *Salsola passerina* Bunge.

**Acknowledgments:** Our work about *Nostoc flagelliforme* has been supported by the Fundamental Research Funds for the Central Universities (No. CCNU16A02007) and the grant from Hubei Key Laboratory of Genetic Regulation and Integrative Biology (2016–2017). We would like to thank the anonymous reviewers for their careful reading and helpful comments on this manuscript.

**Author Contributions:** Xiang Gao drafted the manuscript based on discussions with Wenyu Liang. Wenyu Liang, Haiyan Xu, and Shuifeng Ye revised the manuscript. Xiang Gao and Shuifeng Ye took the photos. Wenyu Liang provided some survey data. All authors read and approved the final manuscript.

**Conflicts of Interest:** The authors declare no conflict of interest.

## References

- Novis, P.M.; Whitehead, D.; Gregorich, E.G.; Greenfield, L.G. Annual carbon fixation in terrestrial populations of *Nostoc commune* (Cyanobacteria) from an Antarctic dry valley is driven by temperature regime. *Glob. Chang. Biol.* **2007**, *13*, 1224–1237. [[CrossRef](#)]
- Belnap, J.; Gillette, D.A. Vulnerability of desert biological soil crusts to wind erosion: The influences of crust development, soil texture, and disturbance. *J. Arid Environ.* **1998**, *39*, 133–142. [[CrossRef](#)]
- Liu, Y.D.; Hu, C.X.; Zhang, W.J. *Desert Algae Environmental Biology and Biological Soil Crust Formation and Sand Fixation*; ISBN: 978-7-03-030232-8. Science Press: Beijing, China, 2013. (In Chinese)
- Gao, K.S. Chinese studies on the edible blue-green alga, *Nostoc flagelliforme*: A review. *J. Appl. Phycol.* **1998**, *10*, 37–49. [[CrossRef](#)]
- Gao, X.; Yang, Y.W.; Ai, Y.F.; Qiu, B.S. Quality evaluation of the edible blue-green alga *Nostoc flagelliforme* using a chlorophyll fluorescence parameter and several biochemical markers. *Food Chem.* **2014**, *143*, 307–312. [[CrossRef](#)] [[PubMed](#)]
- Liang, W.Y. *Research Foundation of Proteomics in Nostoc flagelliforme*; ISBN: 978-7-5525-1532-9. Sunshine Press of the Yellow River Publishing & Media Group: Yinchuan, China, 2014. (In Chinese)
- Liu, N.; Zhou, L.; Hauger, J.S. How sustainable is government-sponsored desertification rehabilitation in China? Behavior of households to changes in environmental policies. *PLoS ONE* **2013**, *8*, e77510. [[CrossRef](#)] [[PubMed](#)]
- Wang, Z.J.; Qiu, X.H.; Tang, Z.H.; Shi, H.X.; Zhao, J.J. Analysis on the factors influencing the evolution of desertification in Ningxia of China from 1999 to 2009. *J. Desert Res.* **2013**, *33*, 325–333. (In Chinese)
- Dai, Z.J. Review on the research of *Nostoc flagelliforme*. *J. Ningxia Univ.* **1992**, *13*, 71–77. (In Chinese)
- Jiao, J.Y.; Zhang, Z.G.; Bai, W.J.; Jia, Y.F.; Wang, N. Assessing the ecological success of restoration by afforestation on the Chinese Loess Plateau. *Restor. Ecol.* **2012**, *20*, 240–249. [[CrossRef](#)]
- Su, J.Q.; Li, X.R.; Li, X.J.; He, H.Y.; Jia, R.L.; Gao, Y.P. Response of vegetation herb layer to nitrogen fertilizer in steppe desert. *J. Desert Res.* **2010**, *30*, 1336–1340. (In Chinese)
- Peng, Q.; Qi, Y.C.; Dong, Y.S.; He, Y.T.; Xiao, S.S.; Liu, X.C.; Sun, L.J.; Jia, J.Q.; Guo, S.F.; Cao, C.C. Decomposing litter and the C and N dynamics as affected by N additions in a semi-arid temperate steppe, Inner Mongolia of China. *J. Arid Land* **2014**, *6*, 432–444. [[CrossRef](#)]
- Chen, X.G.; Li, J.P.; Han, Y.J.; Li, Z.J.; Chen, B.D. Vegetation coverage and its relationships with temperature and precipitation in Ningxia in nearly 20 years. *Chin. J. Ecol.* **2007**, *26*, 1375–1383. (In Chinese)
- Liu, B.R.; Yang, Y.; Chen, L. Distribution characteristics of soil labile organic carbon of four typical plant communities in desert steppe of Ningxia. *Acta Agrestia Sin.* **2014**, *22*, 986–990. (In Chinese)
- Zhang, T.; Lu, S.T.; Li, L. Studies on the diversity of rodent populations in desert steppes in the eastern parts of Ningxia. *Chin. J. Zoonoses* **2013**, *29*, 1229–1230. (In Chinese)
- Parmenter, R.R.; Mesch, M.R.; MacMahon, J.A. Shrub litter production in sagebrush-steppe ecosystem: Rodent population cycles as a regulating factor. *J. Range Manag.* **1987**, *40*, 50–54. [[CrossRef](#)]
- Papanastasis, V.P. Restoration of degraded grazing lands through grazing management: Can it work? *Restor. Ecol.* **2009**, *17*, 441–445. [[CrossRef](#)]
- Jiang, Q.; Li, S.B.; Fan, C.; Liu, X.; Wang, N.G. Sandy desertification of land and control countermeasure in Ningxia. *J. Arid Land Res. Environ.* **1999**, *13*, 54–61. (In Chinese)
- Yang, Z.M.; Li, J.L.; Gan, X.Y.; Yang, F.; Qian, Y.R.; Wang, N. Response of quantity characteristics of vegetation to different grazing intensities in Tan-sheep grazing system in Ningxia Province. *Chin. J. Grassl.* **2009**, *31*, 20–25. (In Chinese)
- Zhang, H.; Shi, S.L.; Wang, S.X. Effect of grazing intensities on plant community structure and grassland productivity in desert steppe of Ningxia. *J. Arid Land Res. Environ.* **2012**, *26*, 73–76. (In Chinese)
- He, N.P.; Zhang, Y.H.; Dai, J.Z.; Han, X.G.; Baoyin, T.; Yu, G.R. Land-use impact on soil carbon and nitrogen sequestration in typical steppe ecosystems, Inner Mongolia. *J. Geogr. Sci.* **2012**, *22*, 859–873. [[CrossRef](#)]
- Shan, G.L.; Chen, G.; Ning, F.; Ma, Y.B.; Chu, X.H. Dynamics of soil microorganism and enzyme activity in typical steppe of restoration succession process. *Acta Agrestia Sin.* **2012**, *20*, 292–297. (In Chinese)
- Hu, R.; Wang, X.P.; Zhang, Y.F.; Pan, Y.X.; Shi, W.; Jin, Y.X. Impact of sand-stabilizing shrubs on soil enzyme activities in Shapotou area. *J. Lanzhou Univ. (Nat. Sci.)* **2015**, *51*, 676–682. (In Chinese)

24. Hong, Y. Preliminary Research on the Ecology of the Soil Algae in Qaidam Basin. Master Thesis, Institute of Hydrobiology, the Chinese Academy of Sciences, Wuhan, China, 1987.
25. Hui, Y.Q.; Zhang, J.W. Analysis of the soil factors for existing *Nostoc flagelliforme*. *J. Gansu Agric. Univ.* **1993**, *28*, 380–383. (In Chinese)
26. Yu, H.F. Effect of mixed carbon substrate on exopolysaccharide production of cyanobacterium *Nostoc flagelliforme* in mixotrophic cultures. *J. Appl. Phycol.* **2012**, *24*, 669–673. [[CrossRef](#)]
27. Ding, Z.; Jia, S.R.; Han, P.P.; Yuan, N.N. Effects of carbon sources on growth and extracellular polysaccharide production of *Nostoc flagelliforme* under heterotrophic high-cell-density fed-batch cultures. *J. Appl. Phycol.* **2013**, *25*, 1017–1021. [[CrossRef](#)]
28. Zhao, H.L.; Guo, Y.R.; Zhou, R.L.; Drake, S. The effects of plantation development on biological soil crust and topsoil properties in a desert in Northern China. *Geoderma* **2011**, *160*, 367–372. [[CrossRef](#)]
29. Liu, Y.D. *Integrated Management of Desertification by “Algae-Grass-Shrub (Tree)” Technology and Regional Sustainable Development*; ISBN: 978-7-03-030246-5. Science Press: Beijing, China, 2013. (In Chinese)
30. Lan, S.B.; Zhang, Q.Y.; Wu, L.; Liu, Y.D.; Zhang, D.L.; Hu, C.X. Artificially accelerating the reversal of desertification: Cyanobacterial inoculation facilitates the succession of vegetation communities. *Environ. Sci. Technol.* **2014**, *48*, 307–315. [[CrossRef](#)] [[PubMed](#)]
31. Melillo, J.M.; Butler, S.; Johnson, J.; Mohan, J.; Steudler, P.; Lux, H.; Burrows, E.; Bowles, F.; Smith, R.; Scott, L.; et al. Soil warming, carbon-nitrogen interactions, and forest carbon budgets. *Proc. Natl. Acad. Sci. USA* **2011**, *108*, 9508–9512. [[CrossRef](#)] [[PubMed](#)]
32. Gao, K.S.; Ye, C.P. Culture of the terrestrial cyanobacterium, *Nostoc flagelliforme* (Cyanophyceae), under aquatic conditions. *J. Phycol.* **2003**, *39*, 617–623. [[CrossRef](#)]
33. Gao, X.; Yang, Y.W.; Cui, L.J.; Zhou, D.B.; Qiu, B.S. Preparation of desiccation-resistant aquatic-living *Nostoc flagelliforme* (Cyanophyceae) for potential ecological application. *Microb. Biotechnol.* **2015**, *8*, 1006–1012. [[CrossRef](#)] [[PubMed](#)]



© 2016 by the authors; licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC-BY) license (<http://creativecommons.org/licenses/by/4.0/>).