

Soil Fungal Community Structure and Diversity in Tamarix Shrublands in the Lower Reaches of the Tarim River (Postprint)

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Abstract

Soil fungal communities play a crucial role in maintaining soil ecosystem functions in arid regions. To investigate the effects of soil physicochemical properties of Tamarix sand mounds and non-sand mounds on soil fungal community structure in the arid region of Xinjiang, this study collected soils from three positions (inside the canopy, canopy edge, and shrubland edge) in Tamarix shrublands with and without sand mounds near the Yingsu cross-section in the lower reaches of the Tarim River. Based on high-throughput sequencing, a preliminary investigation was conducted on the soil fungal community structure and function in Tamarix shrublands with and without sand mounds, and the combined effects of sand mound and soil factors on soil fungal community structure and function were analyzed in conjunction with soil physicochemical properties. The results demonstrated: (1) Soil pH, available potassium, total potassium, ammonium nitrogen, and available phosphorus exhibited significant differences among different positions within the Tamarix shrubland, whereas soil water content, electrical conductivity, total salt, organic matter, total nitrogen, total phosphorus, and nitrate nitrogen showed no significant differences throughout the Tamarix shrubland. (2) Soil fungi in the Tamarix shrubland of this region were classified into 1 kingdom, 14 phyla, 48 classes, 110 orders, 227 families, 410 genera, and 557 species. At the phylum level, Ascomycota, Basidiomycota, and Mortierellomycota were the dominant phyla in this region's Tamarix shrubland; at the genus level, *Alternaria*, *Aspergillus*, *Stolonocarpus*, *Colletotrichum*, *unidentified_{Saccharomycetales}_{sp}*, and *Gymnoascus* were the main dominant genera. (3) Analysis of the relationship between soil physicochemical factors and soil fungal communities revealed that total nitrogen, available potassium, and ammonium nitrogen were the primary environmental factors influencing soil fungal community structure. Total phosphorus showed significant positive correlations with *Aspergillus*, *Microthelia*, *Gymnoascus*, and *Phialosimplex*, while total nitrogen showed a significant positive correlation

with *Alternaria*. (4) Based on FUNGuild fungal functional prediction, three trophic types (saprotrophic, symbiotic, and pathogenic) and five cross-trophic functional guilds were detected in the Tamarix shrubland. Among these, saprotrophic functional fungi (30.0%) dominated the Tamarix shrubland, followed by pathogenic-saprotrophic-symbiotic trophic type (10.6%), pathogenic-symbiotic trophic type (5.9%), and symbiotic trophic type (4.3%), which occupied certain advantages in the Tamarix shrubland. (5) The study found that Tamarix sand mounds and canopies had no significant enrichment effect on soil nutrients and soil fungi; however, functional fungi inside the sand mound canopy showed significant differences from other groups, indicating that the combined effects of Tamarix shrubland sand mounds and canopies exert a substantial influence on soil fungal functional composition.

Full Text

Community Structure and Diversity of Soil Fungi in Tamarix Shrubs in the Lower Reaches of the Tarim River

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Abstract

Soil fungal communities play a crucial role in maintaining soil ecosystem functions in arid regions. To investigate the effects of soil physicochemical properties on soil fungal community structure in nebkha and non-nebkha Tamarix shrubs in Xinjiang's arid region, this study collected soils from three positions (within the canopy, at the canopy edge, and at the shrub edge) in both nebkha and non-nebkha Tamarix shrub communities near the Yingsu section of the lower Tarim River. High-throughput sequencing was employed to characterize the soil fungal community structure and function, and the combined effects of nebkhas and soil factors on soil fungal communities were analyzed in conjunction with soil physicochemical properties. The results showed that: (1) Soil pH, total potassium, available potassium, ammonium nitrogen, and available phosphorus differed significantly among different positions within Tamarix shrubs, whereas soil water content, electrical conductivity, total salt, organic matter, total nitrogen, total phosphorus, and nitrate nitrogen showed no significant differences across the entire shrub system. (2) The soil fungi in Tamarix shrubs were classified into 1 kingdom, 14 phyla, 48 classes, 110 orders, 227 families, 410 genera, and 557 species. At the phylum level, Ascomycota, Basidiomycota, and Mortierellomycota were the dominant phyla, while at the genus level, *Alternaria*, *Aspergillus*, *Stolonocarpus*, *Colletotrichum*, unidentified_{Saccharomycetales}_{sp}, and *Gymnoascus* were the main dominant gen-

era. (3) Analysis of the relationship between soil physicochemical factors and soil fungal communities revealed that total nitrogen, available potassium, and ammonium nitrogen were the primary environmental factors affecting soil fungal community structure. Total phosphorus showed significant positive correlations with *Aspergillus*, *Microthelia*, *Gymnoascus*, and *Phialosimplex*, while total nitrogen was significantly positively correlated with *Alternaria*. (4) Functional fungi dominated the Tamarix shrub communities. (5) Although the enrichment effects of Tamarix nebkhas and canopies on soil nutrients and fungi were not obvious, significant differences in functional fungi were observed within nebkha canopies, indicating that the combined effects of nebkhas and canopies substantially influence the functional composition of soil fungi.

Keywords: high-throughput sequencing; functional gene prediction; nebkhas; soil fungal community; *Tamarix chinensis*

1 Materials and Methods

1.1 Study Area

The study area was located near the Yingsu section in the lower reaches of the Tarim River (87°51' -87°52' E, 40°28' -40°30' N) at an altitude of 830-850 m. This region experiences an extremely arid temperate continental climate characterized by drought, low precipitation, and strong evaporation. Annual precipitation ranges from 17.4 to 42.0 mm, while annual evaporation reaches 2671.4-2902.2 mm. The average annual temperature is 10.6-11.5°C, with monthly averages of 20-30°C in summer (maximum 43.6°C) and -10 to -20°C in winter (minimum -27.5°C). Sunshine duration is 2780-2980 hours, total solar radiation is 5692-6360 MJ · m⁻², frost-free period is 187-214 days, and maximum wind speed is 20-40 m · s⁻¹. The main tree species is *Populus euphratica* Oliv., dominant shrubs include *Tamarix* spp. and *Halostachys caspica*, and major herbaceous plants include *Alhagi sparsifolia* and *Glycyrrhiza uralensis*, with Tamarix being the dominant shrub species.

1.2 Experimental Design and Sample Collection

In October 2019, soil samples were collected from Tamarix communities in the lower Tarim River. Within the sampling plots, three nebkha Tamarix shrubs (approximately 3 m × 3 m) and three non-nebkha Tamarix shrubs of similar size were selected. Using the plant base within the canopy as the center, soil samples were collected radially from three positions: within the canopy (0-50 cm from the plant base), at the canopy edge (50-100 cm), and at the shrub edge (100-150 cm), with 0.5 m spacing between positions. After removing the dry surface soil (0-20 cm), five soil samples were collected from the 20-40 cm layer in the four cardinal directions (east, south, west, north) at each position and mixed. The mixed samples were then reduced using the quartering method.

Each sample was divided into three portions: the first was placed in an ice box, weighed immediately for fresh weight, and then oven-dried for soil water content determination; the second was air-dried indoors, passed through a 2 mm sieve, and used for physicochemical analysis; the third was stored in liquid nitrogen for DNA extraction.

1.3 Measurement Methods

Soil physicochemical properties were determined using Bao Shidan's methods [28]. Specific soil parameters, measurement methods, and instruments are listed in . DNA extraction was performed using the $\{\text{METHOD}\}$ method. Sample purity and concentration were checked via agarose gel electrophoresis. Appropriate samples were selected and amplified using specific primers ITS5-1737F (5' -GGAAGTAAAAGTCGTAACAAGG-3') and ITS2-2043R (5' -GCTGCGTTCTTCATCGATGC-3') with the Phusion® High-Fidelity PCR Master Mix with GC Buffer. PCR products were detected via electrophoresis, and target bands were recovered using Qiagen's gel extraction kit. The TruSeq® DNA PCR-Free Sample Preparation Kit was used for library construction. Qualified libraries were quantified with Qubit and sequenced on the NovaSeq6000 platform (Beijing Compass Biotechnology Co., Ltd.).

1.4 Data Analysis

After sequencing, barcodes were removed and paired-end reads were merged using FLASH [17]. Quality control and chimera removal were performed with Qiime [18] to obtain effective tags. Sequences were clustered into operational taxonomic units (OTUs) at 97% similarity using Uparse [19]. Species annotation was performed with the UNITE database [20] to obtain taxonomic information at all levels. Alpha diversity indices (Chao1, Shannon-Wiener, Simpson, and coverage) were calculated. NMDS (nonmetric multidimensional scaling) and db-RDA (distance-based redundancy analysis) were conducted in R (Version 2.15.3). Fungal functions were predicted by comparison with FUNGuild. Differences in soil physicochemical properties and fungal alpha diversity between nebkha and non-nebkha shrubs were analyzed using t-tests and one-way ANOVA with pairwise comparisons.

2 Results

2.1 Soil Physicochemical Properties

Soil pH in the study area ranged from 7.5 to 8.0, indicating weakly alkaline soils. Soil pH was significantly higher in nebkha than in non-nebkha shrubs, but showed no significant differences among positions within shrubs. Electrical conductivity, total salt, total phosphorus, total nitrogen, available potassium, available phosphorus, and nitrate nitrogen all showed maximum values within

the canopy in both nebkha and non-nebkha shrubs. Available potassium content was significantly higher than other nutrients, decreasing from inside to outside the shrubs, with significant differences among positions in nebkha shrubs but not between nebkha and non-nebkha shrubs. Total potassium content was significantly greater in nebkha than non-nebkha shrubs and differed significantly among positions in non-nebkha shrubs. Ammonium nitrogen showed maximum values at the edge of non-nebkha shrubs, significantly higher than other positions within these shrubs, and was significantly greater in non-nebkha than nebkha shrubs overall. Available phosphorus was significantly higher inside than outside the canopy but did not differ between nebkha and non-nebkha shrubs. Soil water content, total salt, electrical conductivity, organic matter, total nitrogen, total phosphorus, and nitrate nitrogen showed no significant differences across Tamarix shrubs (Table 2).

2.2 Alpha Diversity of Soil Fungi

Coverage values exceeded 99% for all samples, indicating sufficient sequencing depth. Shannon-Wiener index, Simpson index, and Chao1 index showed no significant differences between nebkha and non-nebkha Tamarix shrubs, suggesting similar fungal abundance, diversity, and evenness (Table 3).

2.3 Species Composition of Soil Fungal Communities

A total of 557 species were obtained, with 62.99% successfully annotated in the UNITE database. At different taxonomic levels, 14 phyla, 48 classes, 110 orders, 227 families, 410 genera, and 557 species were detected. At the phylum level (Fig. 1), Ascomycota dominated with an average relative abundance of 65.5%, followed by Basidiomycota (18.35%) and Mortierellomycota (10.6%). Ascomycota reached its maximum relative abundance within nebkha canopies (80.5%) and minimum at non-nebkha shrub edges (49.8%). At the genus level (Fig. 2), *Alternaria*, *Aspergillus*, *Stolonocarpus*, *Colletotrichum*, unidentified_{Saccharomycetales}_{sp}, and *Gymnoascus* were dominant, with average relative abundances of 21.1%, 11.5%, 10.6%, 5.3%, 4.1%, and 3.9%, respectively. Other abundant genera (>1%) included *Plectosphaerellaceae*_{unidentified}, *Neomicrosphaeropsis*, *Microthelia*, *Mortierellaceae*_{unidentified}, *Thielavia*, and *Phialosimplex*. Dominant genera varied among groups: *Aspergillus* dominated within nebkha canopies and edges (25.3% and 11.5%, respectively), while *Plectosphaerellaceae*_{unidentified} dominated nebkha shrub edges (11.5%). In non-nebkha shrubs, *Alternaria*, *Stolonocarpus*, and unidentified_{Saccharomycetales}_{sp} were dominant at canopy interior, edge, and shrub edge positions, respectively.

2.4 NMDS Analysis of Soil Fungal Communities

NMDS analysis based on Bray-Curtis distance (stress value = 0.081) indicated reasonable data reliability. Nebkha shrub samples were more dispersed than non-nebkha samples, suggesting greater internal variation in fungal community

structure within nebkha shrubs. Overall, samples clustered primarily in the first and second quadrants, indicating similarity in soil fungal community structure between nebkha and non-nebkha shrubs (Fig. 3).

2.5 Effects of Soil Physicochemical Properties on Fungal Communities

Distance-based redundancy analysis (db-RDA) revealed that total nitrogen ($R^2 = 0.359$, $P = 0.002$), available potassium ($R^2 = 0.347$, $P = 0.018$), and ammonium nitrogen ($R^2 = 0.581$, $P = 0.032$) were significantly correlated with fungal community structure and were the main environmental factors influencing community composition (Fig. 4). The first two axes explained 38.60% and 20.25% of the variance, respectively. Spearman correlation analysis between the top 30 genera and environmental factors showed that *Aspergillus* was significantly positively correlated with total phosphorus, available potassium, electrical conductivity, and total salt ($P < 0.01$) but negatively correlated with total potassium ($P < 0.05$). *Microthelia* was significantly positively correlated with organic matter and total phosphorus ($P < 0.01$) and with available phosphorus ($P < 0.05$) but negatively correlated with soil water content ($P < 0.01$). *Gymnoascus* was significantly positively correlated with total phosphorus ($P < 0.05$), while *Alternaria* was significantly positively correlated with total nitrogen ($P < 0.05$). *Phialosimplex* showed significant positive correlations with total phosphorus ($P < 0.01$) and available potassium ($P < 0.05$) but significant negative correlation with total potassium ($P < 0.01$) (Fig. 5).

2.6 Functional Prediction of Soil Fungal Communities

Based on FUNGuild analysis, Tamarix shrub soil fungal communities included three main trophic types (saprotrophic, symbiotrophic, and pathotrophic) and five cross-trophic functional groups (pathotrophic-symbiotrophic, pathotrophic-saprotrophic, pathotrophic-symbiotrophic-saprotrophic, saprotrophic-symbiotrophic, and undefined). Saprotrophic fungi dominated (30.0%), followed by symbiotrophic fungi (10.6%). Pathotrophic fungi harmful to plants had low average relative abundance. Saprotrophic and symbiotrophic fungi were more abundant in nebkha than non-nebkha shrubs and higher within canopies than at other positions. Saprotrophic and symbiotrophic fungi were significantly higher within nebkha canopies than other groups, while pathotrophic-symbiotrophic fungi were significantly higher within non-nebkha canopies. Clustering analysis showed that nebkha and non-nebkha shrub edges clustered together first, followed by canopy edges, while nebkha canopy interiors formed a distinct cluster, indicating unique functional group composition (Figs. 6 and 7).

3 Discussion

Spatial heterogeneity of soil resources is common in arid and semiarid regions, and the enrichment of soil nutrients and salts by shrub nebkhas is an important mechanism underlying this heterogeneity. In this study, except for pH, total potassium, and ammonium nitrogen, most soil factors showed no significant differences between nebkha and non-nebkha Tamarix shrubs, possibly because the selected nebkhas were relatively young and had not yet developed obvious enrichment effects. However, organic matter, total nitrogen, total phosphorus, available potassium, electrical conductivity, and total salt all showed maximum values within canopies, with higher values in nebkha than non-nebkha canopy interiors. This suggests that besides nebkha effects, canopies themselves can accumulate soil nutrients, likely because root exudates containing various inorganic ions, sugars, and amino acids provide additional nutrient and energy sources, while litter decomposition within canopies improves soil conditions.

Alpha diversity analysis revealed no significant differences in soil fungal community diversity and abundance between nebkha and non-nebkha shrubs. NMDS analysis also showed similar fungal community structures between the two shrub types, possibly because soil physicochemical differences were not pronounced or because the overall soil environmental conditions were similar.

At the phylum level, Ascomycota dominated, consistent with many studies in arid regions. Ascomycota contains numerous saprotrophic fungi capable of decomposing lignified plant debris. Its highest abundance within nebkha canopies may result from nutrient enrichment creating suitable conditions for these fungi to utilize degradable litter, promoting rapid growth and reproduction. At the genus level, *Alternaria*, *Aspergillus*, *Stolonocarpus*, *Colletotrichum*, unidentified_{Saccharomycetales}_{sp}, and *Gymnoascus* were dominant, all belonging to Ascomycota, similar to findings in desert sand-fixing plant rhizospheres. *Alternaria* and *Colletotrichum* are pathogenic genera that can harm plants, while *Aspergillus* is widely distributed and some species produce enzymes that degrade lignin.

Correlation analysis showed that total nitrogen, available potassium, and ammonium nitrogen were the main factors affecting fungal community structure. Different dominant genera showed varying responses to soil factors, likely due to differences in physiological characteristics and ecological preferences. Functional prediction revealed that saprotrophic fungi dominated Tamarix shrub communities, while pathotrophic fungi were relatively low, indicating a healthy fungal community structure. Functional groups differed significantly among positions, particularly within nebkha canopies, demonstrating that the combined effects of nebkhas and canopies substantially influence fungal functional composition. However, 17.6%-53.2% of fungi remained unannotated, warranting further functional studies.

4 Conclusions

This study investigated soil fungal community structure and function in nebkha and non-nebkha *Tamarix* shrubs in the lower Tarim River using high-throughput sequencing. The main conclusions are:

- 1) *Tamarix nebkhas* and canopies had some enrichment effects on soil nutrients, salts, and fungal abundance, but these effects were not significant for most soil factors.
- 2) A total of 557 fungal species were identified, with Ascomycota, Basidiomycota, and Mortierellomycota as the dominant phyla, and *Alternaria*, *Aspergillus*, and *Colletotrichum* as the dominant genera.
- 3) Total nitrogen, available potassium, and ammonium nitrogen were the primary environmental factors influencing soil fungal community structure. Total phosphorus was significantly positively correlated with *Aspergillus*, *Microthelia*, *Gymnoascus*, and *Phialosimplex*, while total nitrogen was significantly positively correlated with *Alternaria*.
- 4) Three main trophic types (saprotrophic, symbiotrophic, and pathotrophic) and five cross-trophic functional groups were detected, with saprotrophic fungi dominating the community.

Future studies should examine older nebkhas and different soil depths to better understand soil microbial communities and provide more scientific data for ecological restoration in the Tarim River basin.

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References

- [1] Zou Quancheng, Tang Feifei, Liu Zhongyuan, et al. A novel calcineurin B like proteins gene (ThCBL4) improving the salt tolerance in *Tamarix hispida* by transient overexpression[J]. Forest Research, 2018, 31(3): 63-70.
- [2] Ning Husen, He Miao, Luo Qinghong, et al. Evaluation of ecosystem services of *Tamarix chinensis* forest in Xinjiang[J]. Ecological Science, 2019, 38(4): 111-118.
- [3] Tan Fengzhu, Wang Xueqin, Wang Haifeng, et al. Wind tunnel simulation on distribution change of erosion and deposition around nebkhas and interdune under different background vegetation coverage[J]. Arid Land Geography, 2018, 41(1): 56-65.
- [4] Liu Bo, Liu Hongling, Mu Yudi, et al. Correlation between the stable carbon isotopes in annual layers of *Tamarix ramosissima* hillocks in the lower reaches of the Tarim River[J]. Arid Zone Research, 2018, 35(3): 728-734.

- [5] Li Jun, Zhao Chengyi, Zhu Hong, et al. Species effect of *Haloxylon ammodendron* and *Tamarix* spp. on shrub fertile islands[J]. *Acta Ecologica Sinica*, 2008, 27(12): 5138-5147.
- [6] Xing H Q, Xiao Z W, Yan J Z, et al. Effects of continuous cropping of maize on soil microbes and main soil nutrients[J]. *Pratacultural Science*, 2011, 28(10): 1777-1780.
- [7] Han Y Z, Zeng B, Huang J G. Studies on Italian ryegrass rhizosphere microbes[J]. *Chinese Journal of Grassland*, 2011, 33(4): 21-28.
- [8] Wei X R, Huang M B, Shao M A, et al. Shrubs increase soil resources heterogeneity along semiarid grass slopes in the Loess Plateau[J]. *Journal of Arid Environments*, 2013, 88(1): 175-183.
- [9] Cao C Y, Abulajiang, Yusuwaji, Zhang Y, et al. Assessment of the effects of phytogenic nebkhas on soil nutrient accumulation and soil microbiological property improvement in semi arid sandy land[J]. *Ecological Engineering*, 2016, 91: 582-589.
- [10] Chen Ming, Zhu Jianwen, Sheng Jiandong, et al. The effect of *Tamarix* spp canopy on the soil enzyme activities and the microbial quantity[J]. *Acta Agriculturae Boreali Occidentalis Sinica*, 2008, 17(2): 212-217.
- [11] Li Xiqian, Zhang Yuanming. Grey relation analysis on soil fertility as influenced by edge effects of moss crust patch in a temperate desert[J]. *Journal of Desert Research*, 2019, 39(3): 17-24.
- [12] Chen Hongyang, Shang Zhenyan, Fu Hua, et al. Soil microbial biomass and activity under desert shrub canopies[J]. *Acta Prataculturae Sinica*, 2015, 24(2): 70-76.
- [13] Tang Haoqi, Zhang Na, Sun Bo, et al. Effect of interaction between arbuscular mycorrhizal fungi and rhizosphere bacteria in farmland soils on nutrients utilization[J]. *Acta Microbiologica Sinica*, 2020, 60(6): 1117-1129.
- [14] Peng Yujie, Cheng Nan, Li Jiajia, et al. Effects of nitrogen fertilizer reduction on root exudates of maize seedling analyzed by rhizosphere metabolomics[J]. *Chinese Journal of Eco Agriculture*, 2018, 26(6): 21-28.
- [15] Pang Zhiqiang, Yu Diqu. Plant root system microbial interaction system under drought stress and its application[J]. *Plant Physiology Communications*, 2020, 56(2): 109-126.
- [16] Sun Qian. Fungal community diversity and structure in rhizosphere soil of different crops in the arid zone of central Ningxia[J]. *Microbiology*, 2019, 46(11): 2963-2972.
- [17] Mago T, Salzberg S L. FLASH: Fast length adjustment of short reads to improve genome assemblies[J]. *Bioinformatics*, 2011, 27(21): 2957-2963.

- [18] Caporaso J G, Kuczynski J, Stombaugh J, et al. QIIME allows analysis of high throughput community sequencing data[J]. *Nature Methods*, 2010, 7(5): 335-336.
- [19] Rognes T, Flouri T, Nichols B, et al. VSEARCH: A versatile open source tool for metagenomics[J]. *Peerj*, 2016, 4(10): 1-22.
- [20] Haas B J, Gevers D. Chimeric 16S rRNA sequence formation and detection in Sanger and 454 pyrosequenced PCR amplicons[J]. *Genome Research*, 2011, 21(3): 494-504.
- [21] Kõljalg U, Nilsson R H, Abarenkov K. Towards a unified paradigm for sequence based identification of fungi[J]. *Molecular Ecology*, 2013, 22(21): 5271-5277.
- [22] de Boer W, Folman L B, Summerbell R C, et al. Living in a fungal world: Impact of fungi on soil bacterial niche development[J]. *Fems Microbiology Reviews*, 2005, 29(4): 795-811.
- [23] Guo Chengjin, Zhang Lirong, Shen Ruiqing, et al. Diversity of rhizosphere soil fungi in sand fixation plants in Tengger Desert of Ningxia Autonomous Region[J]. *Mycosystema*, 2017, 36(5): 552-562.
- [24] Kang Ziteng, Jiang Liming, Luo Yiyong, et al. The research advances of mechanism of pathogenicity of *Alternaria* phytopathogenic fungi[J]. *Chinese Bulletin of Life Sciences*, 2013, 25(9): 908-914.
- [25] Wu Liangqing, Zhu Liwu, Heng Wei, et al. Identification of Dangshan pear anthracnose pathogen and screening fungicides against it[J]. *Scientia Agricultura Sinica*, 2010, 43(18): 3750-3758.
- [26] Yu Hongyan, Zeng Guangming, Huang Guohe, et al. Screening of lignin degrading fungi and their enzyme production[J]. *Chinese Journal of Applied & Environmental Biology*, 2004, 10(5): 639-642.
- [27] Ma Jian, Liu Xiande, Li Guang, et al. Evaluation on soil fertility quality of *Picea crassifolia* forest in middle Qilian Mountains[J]. *Arid Land Geography*, 2019, 42(6): 1368-1377.
- [28] Bao Shidan. *Soil and agricultural chemistry analysis*[M]. 3rd ed. Beijing: China Agriculture Press, 2000.

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