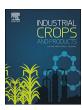
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## Competitive characteristics related to nitrogen utilization and calla lily growth in rubber–calla lily intercropping systems<sup>★</sup>



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#### ABSTRACT

In this study, we analyzed the interspecific competitive relationships and their effect on the growth and nitrogen (N) utilization characteristics of calla lilies in rubber–calla lily intercropping systems using root barrier and <sup>15</sup>N tracer techniques. Our results showed that intercropping significantly inhibited calla lily growth and reduced nitrogen uptake. Nitrogen utilization efficiency was also reduced in the individual parts of calla lily plants, as well as on a whole-plant basis, as a result of competition with rubber trees for nitrogen. Furthermore, nitrogen uptake by calla lily from fertilizer was lower than that from soil, and the percentage of nitrogen taken up from the fertilizer and the nitrogen utilization efficiency calculated by the tracer method were lower than those calculated using the difference method. Our results demonstrated that there two ways in which interspecific competition for nutrients between calla lilies and rubber trees can be mitigated: (1)calla lilies use adaptive strategies (such as increased root length, increased root numbers, and enhanced dry matter allocation to roots) to maximize their nutrient access and (2) N fertilizer application.

#### 1. Introduction

In order to successfully intercrop rubber plantations, increase output, and provide employment opportunities and income for rubber farmers, it is crucial that the land under mature rubber plantations, which can reach up to millions of hectares, is developed. The agronomic benefits of intercropping include improved land and solar radiation utilization, improved retention of soil moisture, improved soil fertility, and decreased erosion in wet and sloping areas (Rodrigo et al., 2001a,b; Guo et al., 2006; Rodrigo et al., 1997; Battany and Grismer, 2000).

China has nearly one million hectares of rubber plantations, and mature rubber plantations account for 70% of total rubber plantations. Currently, rubber plantation intercropping is primarily used in young rubber plantations and is seldom used in mature rubber plantations (Rodrigo et al., 2005; Pathiratna and Perera, 2006). The development of mature rubber plantation intercropping has largely been unrealized for two reasons. First, common agricultural field crops cannot be cultivated in the shade of mature rubber trees. Second, intense nutrient competition between crops prevents successful intercropping. Competition for nitrogen is especially severe because mature rubber trees have large root systems that allow them to monopolize the available nitrogen. Indeed, studies have found that interspecific competition is common when two crops are grown together (Vandermeer, 1989;

Celette et al., 2009). Such competition usually decreases survival, growth, and reproduction in at least one of the species (Crawley, 1997). It is thus crucial that the above two issues are addressed in order to increase land utilization rates under mature rubber plantations.

The nutrient demands, sources, and periods indifferent crops vary according to their physiological and biochemical characteristics (Li, 2001; Wang, 2006). Insufficient soil nutrient supplies inevitably lead to excessive competition for soil nutrients, resulting in soil nutrient depletion and declines in soil fertility (Li et al., 2014, 2015). Therefore, in order to decrease competition for soil nutrients, achieve soil nutrient balance, maintain soil function, and protect soil quality, it is crucial to fully understand the nitrogen uptake and utilization characteristics of the intercrops in intercropping systems. This will allow for a more scientific application of fertilizer techniques in intercropping systems.

Mature rubber intercropping in Hainan requires scientifically determined fertilizer rates in order to improve fertilizer utilization efficiency, as Hainan consistently experiences high temperatures and rainy weather that lead to significant fertilizer loss (Liu et al., 1990; Yuan, 2013). Scientific fertilization techniques can prevent inefficient and excessive fertilization, both of which cause significant fertilizer loss, reduced fertilizer utilization, and increased risk of environmental pollution (Mkhabela et al., 2008; Atapattu and Kodituwakku, 2009).

Calla Lily (Alocasia macrorrhiza L.), which is a shade-loving plant

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with good reproduction abilities, extensive growth management, and high tuber starch and sugar contents, is able to grow in mature rubber plantations. The entire calla lily plant can be used for medicinal or ornamental purposes. Rubber–calla lily intercropping systems can produce products such as starch, livestock feed, and medicine.

In rubber–calla lily intercropping systems, calla lilies play a significant role in increasing the output of rubber plantations; however, calla lilies also have a negative impact due to interspecific resource competition, especially interspecific nitrogen competition. Such competition determines whether the rubber–calla lily intercropping system achieves efficient and sustainable production. The main competitive factor in rubber–calla lily intercropping systems is nutrient competition. Therefore, a clearer understanding of nutrient competition, especially nutrient uptake and utilization efficiency of calla lilies, is essential to ensure the efficient use of soil resources in this type of intercropping system; however, the nutrient utilization efficiency of calla lilies remains unclear in the context of rubber–calla lily intercropping systems.

Nitrogen is a nutrient that is required for rubber tree growth, and it is also the mineral element in greatest demand by rubber trees. Nitrogen deficiency decreases the growth, yield, and dry latex content of rubber trees. Moreover, calla lilies require a significant amount of fertilizer, especially nitrogen fertilizer.

In this study, we investigated the effects of intercropping on the growth and nitrogen utilization characteristics of calla lilies in rubber–calla lily intercropping systems using root barrier and <sup>15</sup>N tracer techniques. The findings of this study can be used to facilitate the scientific intercropping of calla lilies in rubber–calla lily intercropping systems, particularly in tropical environments such as Hainan.

#### 2. Materials and methods

#### 2.1. Experimental site

This study was conducted on an experimental farm belonging to the Chinese Academy of Tropical Agricultural Sciences located in Danzhou (109° 28′ 30″ E, 19° 32′ 47″ N; elevation: 114 m), Hainan, China. The soil at the experimental site was a brick-red loam soil with a pH of 6.14, an organic carbon content (C) of 11.83 g kg $^{-1}$ , a total nitrogen content (N) of 0.68 g kg $^{-1}$ , a total phosphorus content (P) of 0.29 g kg $^{-1}$ , a total potassium content (K) of 22.73gkg $^{-1}$ , an available P content of 19.86 mg kg $^{-1}$ , and an available K content of 56.96 mg kg $^{-1}$ . The plough layer (0–20 cm) had an NO3–N content of 7.8 mg kg $^{-1}$  and an NH4 $^+$ -N content of 3.48 mg kg $^{-1}$ at the start of the experiment. The experimental site is located in a hot and humid climate. The mean annual temperature of the area is 23.3 °C, and the annual precipitation is 1826 mm. The area has a distinct rainy season from May to October and a dry season from November to April. The rainy season accounts for 80% of all precipitation. As such, the mature rubber trees cultivated in this area experience a defoliation period from February to March.

#### 2.2. Experimental design

We analyzed the intercropping competition by inserting barriers between the crops to separate the root systems and to effectively remove belowground competition. The root barriers consisted of a 0.15-mm-thickdouble polyethylene film. We placed the root barriers 2 m away from the rubber trees and buried them at a depth of 1.2 m. The experiment consisted of four treatments: Treatment 1 (T1), which included a root barrier without nitrogen fertilizer; Treatment 2 (T2), which included a root barrier with nitrogen fertilizer; Treatment 3 (T3), which had no root barrier or nitrogen fertilizer; and Treatment 4 (T4), which had no root barrier but did include nitrogen fertilizer. All treatments were replicated three times in a randomized design. The treatments with root barriers had no root interspecific competition, and the treatments without root barriers had root interspecific competition.

The goal of this experiment was to compare the agronomic performance and nitrogen utilization characteristics of calla lilies in rubber–calla lily intercropping systems using  $^{15}{\rm N}$  isotope tracer and root barrier techniques. Individual experimental plots measured 60 m² (20 m by 3 m).

#### 2.3. Crop management

Rubber tree (*Hevea brasiliensis* Müll.Arg.) clones (CATAS 8–79) were planted in the year2000 at a density of 476 plants ha $^{-1}$  (7 m  $\times$  3 m). The rubber trees were tapped in 2006.

Calla lily was planted as an intercrop in the available space between the rubber tree rows during July 2014. A distance of 2 m was maintained between the rubber trees and the intercrop strips. The calla lilies were planted at a spacing of  $100~\text{cm} \times 50~\text{cm}$ .

The calla lilies were fertilized with 135 kg ha $^{-1}$  P<sub>2</sub>O<sub>5</sub> applied as superphosphate, 270 kg ha $^{-1}$  K<sub>2</sub>O applied as potassium chloride, and 300 kg ha $^{-1}$ N applied as  $^{15}$ N-labeled urea (10.16 atom % excess, supplied by the Shanghai Chemical Institute, China). Superphosphate and potassium chloride were uniformly applied to the treatments, while the  $^{15}$ N-labeled urea was uniformly applied to each calla lily plant on 30 April 2015.

#### 2.4. Plant sampling and analysis

Ten calla lily plants were sampled in each treatment replicate on 22 October 2015. Plant height (from the tuber base to the tallest leaf) and tuber and root lengths were measured using a ruler, and tuber and root thicknesses were determined using Vernier calipers. The number of leaves and roots per plant were also determined. Each plant was divided into leaf, tuber, and root samples. Each sample was weighed and immediately dried to a constant weight in a forced-air oven at 75 °C. We milled the dried plant samples using a two-stage process to create a fine "floury" consistency. We analyzed the material for nitrogen content and <sup>15</sup>N enrichment in a single determination using a <sup>15</sup>Nisotope mass spectrometer (Isoprime 100/Vario pyrocube, England).

#### 2.5. Calculation method

Our calculation methods were adapted from Zhang et al. (2008) and Meng et al. (2011).

The amount of N taken up by each organ was the dry weight of each organ  $\times$  the N content of each organ.

The amount of N taken up by each plant was the amount of N taken up by each leaf + the amount of N taken up by the tuber + the amount of N taken up by the root.

#### 2.5.1. <sup>15</sup>N tracer method

Using the  $^{15}$ N tracer method, the atom percent excess  $^{15}$ N was the  $^{15}$ N abundance of the sample or the  $^{15}$ N abundance of  $^{15}$ N-labelled fertilizer - the  $^{15}$ N natural abundance (0.366%).

The percentage of nitrogen in the organs derived from fertilizer was the atom percent excess  $^{15}$ N of the organ  $\div$  the atom percent excess  $^{15}$ N of the fortilizer

The amount of N taken up by the organs derived from fertilizer was the amount of N taken up by the organ  $\times$  the percentage of nitrogen in the organ derived from fertilizer.

The amount of N taken up by the plant derived from fertilizer was the amount of N taken up by the leaf derived from fertilizer + the amount of N taken up by the tuber derived from fertilizer + the amount of N taken up by the root derived from fertilizer.

Nitrogen utilization efficiency was the amount of N taken up by the plant derived from fertilizer  $\div$  the N fertilizer application rate  $\times 100\%$ .

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