Soil Air Water

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682

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### **Research Article**

### *Eucalyptus* Trees – *Ageratina adenophora* Complex System: A New Eco-environmental Protection Model

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Eucalyptus trees and Ageratina adenophora complex system, a new ecological phenomenon that is worthy of attention, was proposed firstly in this paper, and some scientific problems were summarized from the new phenomenon. Causes of the new phenomenon were analyzed tentatively from the perspective of ecology. It was pointed out that ecological degradation of Eucalyptus plantation and strong invasiveness of A. adenophora are two apparent reasons for formation of Eucalyptus trees and A. adenophora complex system. Basic view of the authors on causes of the new phenomenon was put forward that resistance unbalance between chemical defensive potential of Eucalyptus trees and chemical invasive potential of A. adenophora might be the fundamental reason for formation of Euclyptus trees and A. adenophora complex system, based on the two apparent reasons respectively connected with allelopathic effects of *Eucalyptus* trees and A. adenophora as dominant species of the complex system. Some proofs from studies on chemical components and biological activities of Eucalyptus species and A. adenophora have proved the basic view of the authors. It was discussed that formation of the complex system would influence some environmental elements such as soil environment, hydrology environment, and biology environment. It was proposed that three key scientific issues, namely chemical mechanism of formation of the complex system, ecological effects of formation of the complex system, and succession trends and impact factors of the complex system should be mainly studied as special researches to probe ecological relationship of exotic species because of absence study on the complex system and objective requirements of production practice in future. It was emphasized that the proposed researches might be of guidance significance to scientific management and sustainable operation of Eucalyptus plantation under the condition of biological invasion.

**Keywords:** Ageratina adenophora; allelopathic effect; complex system; eco-environmental protection; *Eucalyptus* trees

*Received:* November 27, 2012; *revised:* December 15, 2012; *accepted:* February 7, 2013 **DOI:** 10.1002/clen.201200642

### 1 Introduction

It is a hot issue receiving much concern currently that introduced species and exotic species have serious effects on ecological safety and environmental healthy. In China, *Eucalyptus* spp., being typical introduced species, usually originate ecological degradation in their plantation; *Ageratina adenophora*, being the most noxious exotic specie, often causes great damages to plant diversity and ecological safety of invaded regions. It showed that decline of biodiversity is an obvious characteristic of the ecological degradation by us on plant species of *Eucalyptus* plantation in many *Eucalyptus* forestry regions (e.g., Kunming, Chuxiong, Pu'er, etc.) of Yunnan province

**Correspondence:** X. Huang, College of Environmental Science and Engineering, Southwest Forestry University, Kunming 650024, P. R. China in China suggested that little species can survive in *Eucalyptus* plantation while *A. adenophora* can invade and spread in large scale, thus a new ecological phenomenon occurred that is *Eucalyptus* trees and *A. adenophora* complex system (Fig. 1). It is known that *Eucalyptus* species and *A. adenophora* have allelopathic effects on other species, according to other reports [3, 4]. Allelopathic effect has been regarded as an important reason for *Eucalyptus* species repelling other species and biodiversity decreasing in *Eucalyptus* plantation [5]. Also, allelopathic effect has been proved to be a dominant cause for successful invasion of *A. adenophora* [4, 6]. Hereby, many scientific problems can be summarized from emergence of *Eucalyptus* plantation "allow" invasion of *A. adenophora*? What effects does invasion of *A. adenophora* have on *Eucalyptus* plantation? How did *Eucalyptus* trees and

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Figure 1. (a) *E. globulus–A. adenophora* complex system in Kunming, Yunnan, P. R. China; (b) *E. smithii–A. adenophora* complex system in Chuxiong, Yunnan, P. R. China; (c) *E. urophylla–A. adenophora* complex system in Pu'er, Yunnan, P. R. China.

A. adenophora complex system form? Will the two exotic species be mutually beneficial or be mutually exclusive when they get to a new habitant? These issues should be solved in priority for people to recognize ecological relationship of exotic species and protect regional biodiversity. In this paper, causes and effects of *Eucalyptus* trees and *A. adenophora* complex system were analyzed and discussed tentatively from the perspective of ecology, based on formation of the new phenomenon and some scientific problems from it. Moreover, some research proposes for the new phenomenon was put forward that is expected to be helpful to some relative studies on the complex system conducted in future.

### 2 Experimental

#### 2.1 Causes analysis

#### 2.1.1 General understanding

For the formation of complex system of *Eucalyptus* trees and *A. adenophora*, there are two macro explanations. One is ecological degradation of *Eucalyptus* plantation that might create external condition for invasion of *A. adenophora*; the other is strong invasiveness of *A. adenophora* that might provide internal insurance for its spreading.

#### 2.1.2 Ecological degradation of Eucalyptus plantation

Ecological degradation of *Eucalyptus* plantation is mainly presented as soil degradation, productivity declining, biodiversity decreasing. Occurrence of ecological degradation is connected with allelopathic effects of *Eucalyptus* species.

#### (i) Soil degradation & productivity declining

Soil degradation is very common in Eucalyptus plantation in South China. e.g., soil morphology and physicochemical properties degraded seriously in Leizhou Peninsula, Guandong province after cropped Eucalyptus species for several decades [7]. Soil degradation of Eucalyptus plantation is also found in other countries [8-10]. Soil degradation directly leads to productivity declining, which has been proved many times in production practice. Study made in Hetou Forest Farm of Leizhou Forestry Bureau, China suggested that biomass of Eucalyptus plantation decreased with increase of rotations of continuous planting. Taking biomass of single plant for instance, the biomass of single plant of the 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> generation decreased 12.4, 17.2, and 45.0%, compared to the 1<sup>st</sup> generation; taking stock biomass for another instance, the stock biomass of the  $2^{nd}$ ,  $3^{rd}$ , 4<sup>th</sup> generation forest reduced 19.6, 26.7, and 44.6%, compared to the 1<sup>st</sup> generation [11]. Another study showed that productivity potential of Eucalyptus plantation of China should reach 80% of local climatic productivity, but in western Guangdong and Hainan province, the practical productivity of Eucalyptus plantation is 22.8 and 41.1% of the local climatic productivity, respectively [12].

#### (ii) Biodiversity decreasing

More studies on biodiversity problems of Eucalyptus plantation have been reported at home and abroad. e.g., Ramanujam & Kadamban [13] studied dry hot evergreen forestry in Pondicherry area in south India. The result showed that olagapuram grove is heavily degraded as it lost the status of a sacred grove because of its conversion to Eucalyptus plantation. Study by Eshetu & Olavi [2] in E. globulus Labill. sp. globulus plantations in the Ethiopian highlands indicated that indigenous woody species richness and abundance of sample plots at Menagesha where there was remnant natural forest were on average 2.4 and 5.7 times higher, respectively, than the sample plot at Chancho where natural forest were absent. Analysis of plant diversity in Dongmen Eucalyptus plantation of Guangxi, China suggested that continuous cropping of Eucalyptus plantation resulted in a reduction of species diversity. e.g., plant species numbers of the second generation was reduced by 55.43% in a 667 m<sup>2</sup> plot, compared with the first generation [14]. It could be also shown that species and quantities of bacteria, fungi, actinomycetes in pure Eucalyptus plantation is less than in mixed Eucalyptus plantation [15].

(iii) Allelopathic effects of Eucalyptus species

As to causes of ecological degradation of Eucalyptus plantation, different scholars have different viewpoint. However, more and more researchers have recognized that allelopathic effect is a crucial factor that initiated ecological degradation of Eucalyptus plantation. It is showed that leaves, fruits of Eucalyptus species can produce volatile compounds [16-19]. Also, litters decomposition and physiological metabolism of roots can release active chemicals [20, 21]. Accumulation, transportation, conversion of these substances in forest soil might affect physicochemical properties of soil, development of Eucalyptus plantation, which induces species and quantity decline of other organisms [5, 22, 23]. Consequently, Eucalyptus plantation ecosystem might be damaged and its sustainable management might be hindered. It showed that ecological degradation of Eucalyptus plantation is closely related to allelopathic effects of Eucalyptus spp. This would create external condition for invasion of A. adenophora.

#### 2.1.3 Strong invasiveness of A. adenophora

Strong invasiveness of *A. adenophora* can be seen from its basic invasion features. Formation of strong invasiveness of *A. adenophora* is closely relative with its unique biological characteristics and allelopathic effects.

#### 2.1.3.1 Basic features of A. adenophora invasion

Three basic features are presented in A. adenophora invasion.

#### (i) Wide diffusing range

Ageratina adenophora originated in Central American. In recent years, it has invaded and diffused widely in over 30 countries of tropical and subtropical zone [24]. In China, this exotic specie has distributed in each province (autonomous region, municipality) in South-west China and Taiwan province [25]; moreover, it expand to east, north with speed of 20 km per year [26]. Habitats invaded by *A. adenophora* involve farmlands, forests, grasslands and other terrestrial ecosystems, and edge of rivers, lakes, reservoir, wetland and other aquatic ecosystems. This noxious weed even can be found in some nature reserves [27].

#### (ii) Serious invasion damage

Generally, *A. adenophora* invasion leads to regional biodiversity declining or losing [28] and soil quality alteration [29], that endangering regional ecology safety and environment healthy and causing great economic lose and excessively high control cost [30].

#### (iii) Difficulties in prevention and control

At present, no effective technical means can be found to prevent or control *A. adenophora* in many countries. The physical control technique needs huge consumption in human, material, financial resources; the chemical control technique would induce secondary pollution; the biological control technique is regarded as an ideal means for prevention and control of *A. adenophora* but facing some technique difficulties, e.g., how to control and manage the released parasitic insect (e.g., *Procecidochares utilis*), the inoculative virus (e.g., *Alternaria alternate*), the planted species (e.g., *Pennisetum sinese*, *Pennisetum clandestinum*) outdoors to prevent them becoming pests. All these basic features suggested that *A. adenophora* has extraordinary invasive potential.

## 2.1.3.2 Unique biological characteristics of *A.* adenophora

As for the strong invasiveness of A. adenophora, many researchers have made analysis from different perspective of biology. The results showed: Ageratina adenophora has persistent soil seed banks [31]; Ageratina adenophora could acclimate to large extent of environmental light regimes through changes of morphological and physiological characteristics [32, 33]. Ageratina adenophora has evolved into different ecotypes with regard to freezing tolerance through physiological adaptation during their invasion [34]. Ageratina adenophora has both conservative traits and prodigal traits in water utilization [35]. Ageratina adenophora affects growth of local species by changing soil microbial community [29, 36]. Ageratina adenophora appears to have evolved increased N allocation to photosynthesis (growth) and reduced allocation to cell walls, resulting in poorer structural defenses, etc. [37, 38]. These unique biological characteristics and survival strategies are beneficial to A. adenophora adapting to changeable environment, diffusing rapidly, invading successfully [39, 40].

#### (i) Effects of allelopathy in A. adenophora

Doubtlessly, studies mentioned above revealed the strong invasiveness of A. adenophora from different perspective of biology. Nevertheless, more and more researchers tend to explain invasiveness of alien species by using NW (novel weapon) hypothesis [41-44]. NW hypothesis reveals that allelopathy might play dominant roles in invasion of alien species [41]. Actually, studies that revealing invasiveness of A. adenophora from allelopathy has been reported. e.g., Song et al. [4] found that A. adenophora affects other plant species and invades successfully through releasing of volatile allelochemicals from aerial part tissues and leaching of water soluble allelochemicals from both aerial tissues and litters. Study by Yu et al. [6] proved that difference of allelopathy in A. adenophora in various habitats is one cause of its different invasive effect. It is also found that devotion of allelopathy in aerial parts to invasiveness is larger than that of allelopathy in underground parts [6]. Obviously, allelopathy in A. adenophora is a key that forming its strong invasiveness. This provides internal supports for invasion of A. adenophora to Eucalyptus plantation.

#### 2.2 Hypothesis

#### 2.2.1 Basic view of the authors

It seemed to be easy that using the two macro explanation above can analyze the causes of formation of Eucalyptus trees and A. adenophora complex system. However, the key issue lies in: whether do Eucalyptus trees, being allelopathic species, have potential chemical defensive capacity to invasion of A. adenophora? On the contrary, whether does allelopathic invasive potential of A. adenophora exceed chemical defensive capacity of Eucalyptus trees? What interspecific relationship does exist between Eucalyptus trees and A. adenophora? What would be the ecological succession after complex system of Eucalyptus trees and A. adenophora forming? From these questions, it can be seen that the two macro explanations mentioned above (ecological degradation of Eucalyptus plantation and strong invasiveness of A. adenophora) are only apparent reasons for formation of Eucalyptus trees and A. adenophora complex system. They can't be used to reveal essence and mechanism of formation of the complex system. On the contrary, it can be found that there is a common



driving force (or a key factor) that induced ecological degradation of *Eucalyptus* plantation as well as strong invasiveness of *A. adenophora*, according to analysis presented above. It is namely allelopathy. Thereby, it can be inferred that allelopathy might be an important tie between the ecological declines of *Eucalyptus* plantation and the strong invasive capacity of *A. adenophora*. That is to say, *Eucalyptus* trees and *A. adenophora* complex system might originate from allelopathy in *Eucalyptus* species and *A. adenophora*. Deeply speaking, resistance unbalance between chemical defensive potential of *Eucalyptus* trees and *A. adenophora* complex system (Fig. 2). This is the basic view of the authors for formation of *Eucalyptus* trees and *A. adenophora* complex system.

#### 2.2.2 Some relative proofs

The basic view of the authors for formation of the complex system is supported not only by a logical reasoning, but also by some proofs from studies on chemical compounds and biological activities of *Eucalyptus* spp. and *A. adenophora*.

(i) Chemical compounds and biological activities of *Eucalyptus* spp Currently, studies on chemical constituents from *Eucalyptus* spp. are dominated by analysis of essential oil and volatile constituents.
e.g., Batish et al. [16] identified 19 volatile constituents in essential oil from juvenile and senescent leaves of *E. citriodora* compared to 23 in adult leaves and 20 in leaf litter, respectively. Pereira et al. [45] found 33 constituents in essential oil from fruits of *E. globulus*, etc.
[18, 19, 46]. Some chemicals from *Eucalyptus* species had been proved having biological activities. e.g., essential oil from decaying leaves of *E. citriodora* and its major components, citronellal and citronellol, affected germination and root elongation of two weeds (*Cassia occidentalis*, *Echinochloa crus-galli*) [16]; also, essential oil from intact and fallen leaves of *E. citriodora* and its major components (citronellal,



Figure 2. Draft for formation process of *Eucalyptus* trees–*A. adenophora* complex system.

citronellol) were found to be phototoxic against two weeds (Amaranthus viridis, E. crus-galli) and two crops (Triticum aestivum, Oryza sativa) [20]. Study by Singh & Sharma [21] indicated that essential oil from leaves of E. tereticornis Smith and its root extract portioned with ethanol had significant antibacterial activity, etc. [17, 23, 47, 48]. Among them, some studies had proved allelopathy in Eucalyptus species. e.g., aqueous extracts of E. citriodora Hook had greater inhibitory effect on germination and vigour index (shoot length, root length, fresh weight and dry weight) of gram seeds (Cicer arietinum) [49]. Decompose of E. camaldulensis Dehnh leaves decreased catalase activity and increased ascorbate activity in root and shoot of Phalaris in comparison to control while aqueous extracts of Eucalyptus leaves significantly raised ascorbate peroxidase in Phalaris root [50]. Soil from E. grandis, E. urophylla, E. grandis  $\times$  E. urophylla plantation contained inhibitory principles that affected the germination and early growth of crop plants [5, 51, 52].

#### (ii) Chemical compounds and biological activity of A. adenophora

Statistics by Yan et al. [53] showed: 49 compounds were identified in fat-soluble extracts from leaves, stems and inflorescences of A. adenophora, the main chemical structure types of these compounds are sesquiterpenes, steroids and triterpenoids, flavonoids, phenylpropanoids, etc.; 45 compositions were isolated from essential oil of A. adenophora, among these compounds, mass fractions of cymene and bornyl acetate are 20.11 and 12.26%, respectively, which are higher than that of other compositions. Moreover, it was found that some chemical constituents of A. adenophora showing biological activities, e.g., inhibition of 9-beta-hydroxy-phorone, 9-beta-oxoageraphorone from A. adenophora on radicles and coleoptiles growth of wheat (Triticum aestivum), maize (Zea mays L.), E. globulus and other plants [54], toxic activity of euparin A to aphids [55], antifeedant effect of ayapin on animals and insects [56]. Alleopathy in A. adenophora has attracted more attentions of scholars from many countries since 1980s [4, 57-65]. At present, it has been proved that allelpathy effects of this plant play a part in releasing of volatile allelochemicals from aerial part tissues and leaching of water soluble allelochemicals from both aerial tissues and litters [4]. The most active allelochemical in petroleum extract of the aerial part of A. adenophora was identified as 9-oxo-ageraphorone that is a candinene derivative [4]. Two major active allelochemicals were identified in water extract of its aerial part, and they were 4,7-dimethyl-1-(propan-2-ylidene)-1,4,4a,8a-tetrahydronaphthalene-2,6(1H,7H)-dione (DTD) and 6-hydroxyl-5-isopropyl-3,8-dimethyl-4a,5,6,7,8,8a-hexahydronaphthalen-2(1H)-one (HHO) [60]. Furthermore, mechanism of allelopathiic effects of the two allelochemicals DTD and HHO was analyzed. The result showed that seedlings of upland rice treated with the two allelochemicals were stunted; leaves of the seedlings were etiolated and on the leaves surface some physiological speckles appeared [61]; roots of the seedlings presented some symptoms such as being stunted and swollen, and decreasing in amount of lateral roots and nutrient absorption [61]. Appearances and anatomical structures of root tip cells in treated upland rice changed obviously, such as length of cortex parenchyma cells reduced while width increased, and large amount of epidermis cells sloughed off [61]. After exposure to DTD and HHO, malondialdehyde (MDA) content and peroxidase (POD) activity in rice seedlings roots increased significantly, whereas chlorophyl (CHL) content in leaves decreased obviously with prolongation of treatment time and with increasing of allelochemicals concentration (from 0.5 to 1.5 mM),

which suggested that in root cells peroxidation was induced by the two allelochemicals [60, 61]. DTD and HHO also induce accumulation of abscisic acid (ABA) and decrease of indoleacetic acid (IAA) and zeatin ribosides (ZR) in roots of upland rice seedlings with prolongation of treatment time and with increasing of allelochemicals concentration (from 0.5 to 1.5 mM), which would make hormone content *in vivo* becomes unbalanced and seriously affect its growth and development [61–65].

All the above studies proved that *Eucalyptus* species and *A. adenophora* could release active chemicals through various means and have multiple allelopathic effects on other species in different ways and mechanisms. Interactions between *Eucalyptus* species and *A. adenophora* and its occurrences mechanism have not been studied at present, but it is difficulty to find strong evidences or sufficient reasons to deny allelopathic factors that affect coexistences and confrontation of *Eucalyptus* species and *A. adenophora*.

### 3 Results

#### 3.1 Impact analysiss

It might be predicted that formation of *Eucalyptus* trees and *A. adenophora* complex system would have multiple impacts on regional environment. The impacts usually appear in soil environment, hydrology environment and biology environment.

#### 3.1.1 Impact of the complex system on soil environment

Generally, degradation of Eucalyptus plantation leads to soil quality declining [7-10, 15]. However, A. adenophora invasion can improve soil characteristics such as strongly increasing content of nitrate nitrogen (NO<sub>3</sub><sup>-</sup>-N), ammonium nitrogen (NH<sub>4</sub><sup>+</sup>-N), available P and K, and abundance of soil VAM (vesicular-arbuscular mycorrhizal fungi) and the fungi/bacterial ratio that facilitate itself and inhibit natives [29]. It suggested that the impact of the complex system on soil environment is complicated. In the authors' opinion, soil environmental would be positively affected by the complex system in earlier stage of A. adenophora invasion because of increasing vegetation cover and decreasing surface erosion, whereas it would be negatively affected in later period due to huge consumption of soil water and nutrients by more and more A. adenophora population. Certainly, rate and intensity of the impact would depend on comprehensive function of the complex system and all other environmental factors as well as time length of Eucalyptus plantation degradation and A. adenophora invasion.

## 3.1.2 Impact of the complex system on hydrology environment

Before A. adenophora invading, ground vegetation amount in degraded Eucalyptus plantation is limited, that is beneficial to runoff formation and sediment transport. With invasion of A. adenophora, vegetation cover becomes more and more, that effectively reduces surface runoff, soil erosion, and sediment transport. So it can be concluded that hydrology process of surface water would be improved by formation of the complex system. As for hydrology process of soil water, it might be determined by various environmental factors such as rainfall infiltration, surface vegetation, soil texture and characteristic, soil water utilization and storage, etc.

## 3.1.3 Impact of the complex system on biology environment

It could be shown that not only ground biodiversity but also underground biodiversity declines severely after cropping *Eucalyptus* species or during *Eucalyptus* plantation declining [1, 2, 13–15]. Under condition of *A. adenophora* invasion, ground plant biodiversity was damaged ruinously [28] while soil microbial biodiversity was affected positively [29]. From some previous results, it can be deduced that ground biodiversity might become worse and worse after formation of the complex system whereas subsurface biodiversity would restore in earlier years and then decline in later years. Similarly, rate and strength of biodiversity alteration would depend on overall influencing of the complex system and all other environmental factors as well as time length of *Eucalyptus* plantation degradation and *A. adenophora* invasion.

All impacts of formation of the complex system are not immutable but variable with changes of various environmental factors, which indicate the above discussions have no certainty or uniqueness. So it becomes urgent and significant for us to conduct some further researches to verify them.

### 4 Discussion

In the past two decades, ecological issues induced by degraded or damaged ecosystem [2, 9, 10, 66, 67] and exotic species invasions [29, 41, 52, 68, 69] have been recognized by more and more scholars. Some progresses were gotten in research of Eucalyptus plantation degradation as well as A. adenophora invasions from different perspective. This provides some basic references for us to conduct further research in future. However, with expanding scope of ecosystem degradation and exotic invasions, some new things or phenomenon such as Eucalyptus trees and A. adenophora complex system appeared that are great challenging for people to recognize and investigate. From above causes analysis on Eucalyptus trees and A. adenophora complex system, it can be seen that allelopathy in Eucalyptus species and A. adenophora inevitably affect species regeneration and community succession in ecosystem of Eucalyptus plantation. However, research report that is related to occurrence and existence of Eucalyptus trees and A. adenophora complex system has not been found. Accordingly, analysis on chemical interaction between Eucalyptus species and A. adenophora is lack. Besides, current studies on Eucalyptus plantation or A. adenophora is dominated by causes and effects of ecological issues originate from Eucalyptus species and A. adenophora, respectively. There is no research report concerning interaction relationship between Eucalyptus species and A. adenophora and its occurrence mechanism under natural condition or artificial compound system. Based on such research status, moreover, objective requirements of productive practice conducted in future (e.g. operation and management of plantation, prevention and control of exotic species.) should be taken into account, the authors proposed that three key scientific issues below should be mainly studied as special researches in future.

# 4.1 Chemical mechanism on formation of the complex system

This study aims to reveal chemical mechanism of ecological relationship between *Eucalyptus* trees and *A. adenophora* and maintaining or changing of the relationship. It has profound guiding significance to



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No.	Analyzed item	Tested index
1	Soil ecology	
1.1	Soil physical properties	Soil texture, soil bulk density, soil porosity, soil water content, etc.
1.2	Soil chemical properties	Cation exchange capacity (CEC), total amount of mineral, effective nutrient content, organic matter content, acidity and alkalinity, etc.
1.3	Soil biological properties	microbial composition, enzyme activity, etc.
2	Hydrological ecology	
2.1	Rainfall	Rainfall, rainfall intensity, etc.
2.2	Runoff	Runoff, runoff modulus, etc.
2.3	Sediment	Sediment transport rate, sediment transport modulus, etc.
3	Plant ecology	
3.1	Population ecology & community ecology	Richness, evenness, diversity indexes (Simpson's diversity index, Shannon Wiener index), etc.
3.2	Individual ecology	Seedling survival rate, individual density, plant height, biomass (fresh weight, dry weight), etc.
3.3	Physiological ecology	Water utilization efficiency, nutrients (Ň, P, K) utilization rate, chlorophyll content, photosynthetic rate, respiratory rate, etc.

sustainable and scientific management of *Eucalyptus* plantation under the condition of biological invasion. This study can be divided into two parts below.

(i) Chemicals foundation for formation of *Eucalyptus* trees and *A. adenophora* complex system

Chemical constituents of *Eucalyptus* species and *A. adenophora* can be isolated and identified by using modern chemical technologies and means. Portion ratio and distribution status of main chemical components in plant and in soil can be analyzed, too. Major chemical compositions with bioactivity from *Eucalyptus* species and *A. adenophora* can be tested primarily based on bioassay. Main chemical constituents that having adverse effects on *Eucalyptus* species and other plant species in *Eucalyptus* plantation, and those that having positive or negative effects on *A. adenophora*, should be mastered in priority.

(ii) Allelopathic mechanism on formation of *Eucalyptus* trees and *A. adenophora* complex system

Seed culture and seedling planting of *Eucalyptus* species, *A. adenophora* and other plant species in *Eucalyptus* plantation should be conducted with biological stimulation method to set up a system for experimental stimulation study on formation mechanism of *Eucalyptus* trees and *A. adenophora* complex system. The main chemical constituents with biological activities (determined in the first section) should be fertilized in the experimental stimulation system in some way. Allelopathic effects and influencing mechanism of the active chemicals on growth rates, dry matter accumulation, water and fertilizer utilization, photosynthesis, and respiration of plant species cultured or planted in the stimulated system should be tested and analyzed. Furthermore, allelopathic mechanism on formation of *Eucalyptus* trees and *A. adenophora* complex system would be revealed.

# 4.2 Ecological effects of formation of the complex system

With regard to the complex system formed by *Eucalyptus* trees and *A. adenophora*, it is necessary for us to conduct systematic research to investigate its effects on some ecological factors in that multiple mechanisms underlie rapid expansion of an invasive alien plant [68]. In this study, field experimental observation points of *Eucalyptus* 

plantation with different degree of *A. adenophora* invasion should be established to meet study requirement. Based on long-term positioning observation and tracking survey on some ecological factors of the complex system combined with sampling and testing, various possible ecological effects of *A. adenophora* invasion on *Eucalyptus* plantation and ecological characteristics and dynamic changes of the complex system would be determined through analysis of three aspects and some indexes related to the complex system are listed in Table 1 in the following.

# 4.3 Succession trends and impact factors of the complex system

Various environmental factors such as soil, hydrology, and biology would be influenced extensively by formation of the complex system. On the contrary, the complex system would be affected by single feedback or multiple feedbacks of different environmental factors that would underlie succession trends of the complex system respectively or jointly. Therefore, the key study on succession trends of the complex system should be to analyze and grasp feedback effects of various environmental factors on the complex system. For this research, a long-term systematic experiment related to succession trends of the complex system would be designed and carried out in one or more field experimental stations with remarkable differences in many environmental factors such as topography, altitude, natural light, atmospheric temperature, radiant heat, rainfall recharge, soil type, biological interventions, human activities, etc. These environmental factors would be investigated in the systematic experiment to clarify effects of them on community structure and ecological function of the complex system and to determine its succession trends. This study is significant for us to understand influencing mechanism of the environmental factors and succession law of the complex system.

#### Acknowledgments

The authors would like to thank National Natural Science Foundation of China (No. 31160155 & No. 31270751), Applied Basic Research Program of Yunnan province, China (No.2007C022M) and Scientific Researching Project of Southwest Forestry University, Kunming, China (No.110714) for financial support of this work. The authors appreciate Professor GuangJun Zhang from North-West Agriculture and Forestry University, Yangling, China. and Professor YongQing Ma from Institute of Soil and Water Conservation, Chinese Academy of Sciences, Yangling, China. for assisting with the preparation of the revised manuscript. The authors acknowledged the constructive comments and suggestions of anonymous reviewers.

The authors have declared no conflict of interest.

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