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Full Length Article

Studies on the Dynamic Changes in Plant Nutrients Organs and Underground Vegetation of Chinese Fir Plantations

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Abstract

Determination of nutrient contents is important as it provides theoretical and technical support for tending of the Chinese Fir plantation. In this study, dynamic changes in plant nutrient content in Chinese fir [*Cunninghamia lanceolata* (Lamb.) Hook] plantations at different ages were studied. Data on continuous positioning were obtained from a Shangpuzi Woodland Ridge in Suining, Hunan province, China. The results showed that at the same age, different organs of Chinese fir possessed different nutrient content in the following order: needle > twig > bark > root > trunk. The order of each nutrient element was Ca > N > K > Mg > P in bark and root, N > Mg > Ca > K > P in trunk, and Ca > K > N > Mg > P in twig. The nutrient content of plant organs was highest in fast-growing (7 to 12-year old) period, and began to decline when entering the trunk wood stage, though the downward trend differed. The nutrient content in the same organs differed amongst plants of different ages. The variation in nutrient content in the undergrowth vegetation with forest age was comparable to that in the tree layer, whilst the content in the organs exceeded that of the corresponding organs of the tree layer. Upon comparison to the living tree layer, the nutrient content in plant tissues at different growth stages were determined not only by the genetic and physiological characteristics of the plant itself, but further influenced by temperature, precipitation and other habitat conditions.

Keywords: Chinese fir plantations; Continuous positioning; Growth stages; Nutrient contents

Introduction

Plant growth is dependent on nutrient content and balance (Alban 1973). Trees exhibit higher yields when their nutrient composition and distribution are in good balance. Nutrient contents therefore directly determine the productivity of the forest. Moreover, nutrient content can reflect the influence of the surrounding environment on plant growth.

As early as the 1950s, the content of plant nutrient elements were under study (Hou *et al.* 1959). Studies on nutrient content and their distribution in plants increased rapidly in China. For example, significant differences in the nutrient content of Chinese fir [*Cunninghamia lanceolata* (Lamb.) Hook] were observed in different regions (Pan *et al.* 1979; Feng *et al.* 1985). A study on both nutrient content and their distribution in Chinese fir during the fast-growing stages has been conducted (Xiang *et al.* 2002). Similar studies on nutrient content of *Cathaya argyrophylla* (Wang *et al.* 1983; Shen *et al.* 1985), *Eucalyptus urophylla* (Dong *et al.* 1986) and *Acacia mangium* are also reported (Lin *et al.* 2002). These studies have important practical and theoretical significance for the protection and utilization of forests.

In the past, although the nutrient content of different plant organs has been measured, an in-depth analysis of the underlying mechanisms has not been known yet. Nutrient content had been assessed as a measurement parameter, with studies on the accumulation and distribution of nutrients in the trees, nutrient absorption, storage and cycling processes assessed. The dynamics of nutrient content in specific organs at different growth stages had also been assessed (He *et al.* 2007) using the "space-for-time" method. However, due to the heterogeneity of space the environmental conditions differ, leading to uncertainty in the data. In this study, the nutrient content of different organs of Chinese fir at different ages were studied, based on measurement data from 20 years of continuous positioning from a Shangpuzi Woodland Ridge in Suining, Hunan province, China. The data provide important information for nutrient cycling research and nutritional diagnosis for Chinese fir, and provide a basis for the cultivation and management of plantations.

Materials and Methods

Experimental site

This study assessed the artificial fir forest in Shangpuzi Woodland Ridge in Suining, Hunan province. The region is characterized by a mid-subtropical humid monsoon climate,

with an elevation of 190–460 m, an annual average temperature of 16.7°C, and an annual average precipitation of 1320 mm. No temperature extremes occur in either summer or winter. However, the temperature fluctuates between day and night and variations in the climate and vegetation amongst vertical positions are observed. The forest soil was yellow with medium organic matter, and the thickness of the soil layer was ≥ 80 cm. The content of humus in the surface soil was 33.62–41.53 g/kg, whilst the content of total nitrogen (N), phosphorus (P) and potassium (K) were 14.02–21.65, 0.47–0.66 and 14.26–21.83 g/kg, respectively. The forest stand represented a Chinese fir plantation built in reclamation area of the broad-leaved secondary forest in 1990. The afforestation density was 2380–2560 trees/hm^2. In the first 3 years following afforestation, the plantation only thinned during spring and autumn. Abundant undergrowth vegetation was observed for *Maesa japonica*, *Ilex chinensis*, *Pinus albicaulis*, *Litsea cuheha*, *Urena procumbens*, *Herba agrimoniae*, *Woodwardia japornica* and *Houttuynia cordata*.

Sample collection

Sample plots were established for biomass determination (667 m^2) for seven years after building the plantation. Biomass was measured in the plots using the methods proposed by Pan Weijun when the growth stage reached 7, 12, 15, 18, 21 and 25 years (Pan *et al.* 1978).

To guarantee a true representation of the tree samples, we excavated intact standard trees (according to the average tree factor in the plot) with roots, and sampled equal-weight tree trunks per meter, which were uniformly mixed in all sub-samples to obtain an ultimate trunk sample. The sampling methods of twigs, needles, and bark samples were comparable to those of trunk samples. The roots were divided into four components; fine roots (root diameter < 0.2 cm), coarse roots (0.2 cm ≤ root diameter < 1 cm), large roots (root diameter ≥ 1 cm) and root tips. Mixed roots were sampled according their relative weight in the root biomass.

Undergrowth vegetation and litter in the plots were simultaneously collected. Subplots (2 m × 2 m) were established at each of the four corners (~1 m from the boundary of the plot) and at the center. All the undergrowth vegetation in the subplots was collected including leaves, twigs (with small stem also) and root samples. Whole litter in the subplots was also collected, and divided into three components, namely leaves, twigs and debris. The fresh weight of these three components was measured in situ, and mixed according to their relative weight in the fresh litter. In addition, dead root samples in the soil were obtained when the standard trees were excavated.

Determination of nutrient content

Total N and P were measured using the semimicro-Kjeldahl method and molybdenum-blue colorimetry, respectively. Total K, Ca and Mg were determined by atomic absorption spectrophotometer. Measurements were performed five times and mean values calculated. Data were processed on Excel software, and statistical analysis was performed using SPSS 13.0. One-way and two-way ANOVA were used to evaluate the variation amongst different treatments. Duncan Multiple Range test was used to identify significant differences between the treatments.

Results

Nutrient content in the plant organs of Chinese fir

The nutrient content of different organs showed comparable regularity regardless of the fir age in the order: needle > twig > bark > root > trunk ($p< 0.05$; Table 1). The contents of each nutrient element differed in the same plant organ at the same growth stage. The content of nutrient elements was Ca>N>K>Mg>P in bark and roots, N>Mg>Ca>K>P in the trunk, Ca>K>N>Mg>P in the twigs and N>Ca>K>Mg>P in the needles. Duncan tests showed that the differences were significant ($P<0.05$), excluding the content of Mg and Ca in the trunk that did not differ ($P>0.05$) in the fir forest at age of 15, 18, 21 and 25 years. Regardless of the age of the fir, the content of N in each organ were in the order needle > bark > twig > root > trunk; P, K, Ca were needle > twig > bark > root > trunk; whilst those for Mg were in the order needle > twig > root > bark > trunk.

The specific nutrient elements and total nutrients in all plant organs increased from year 7 to year 12-year, but declined at year 15 with differing trends (Table 1). The nutrient content in the plant trunk and bark continued to decline until year 25, whilst that of needles, twigs, and roots declined only to year 21, but augmented at year 25.

Nutrient contents in undergrowth vegetation

Nutrient content in the undergrowth vegetation was more abundant than that of the tree layer (Table 2). The nutrient content in the leaves was highest, followed by twigs, whilst the lowest value was observed in the roots. Meanwhile, the nutrient content in each undergrowth vegetation organ differed, being N>K>Ca>Mg>P in leaf, K>N>Ca>Mg>P in twigs (including small stems), compared to N>Ca>K>Mg>P in the roots. Significant differences in the content of each element were also observed across the different undergrowth vegetation organs at the same growth stages ($p<0.05$).

There were significant differences in the N, Ca and Mg content of the same tissues across the growth stages (Table 2). Nevertheless, the P content showed no significant differences across the growth stages, except for 7-year olds, which were significantly lower ($p<0.05$). There were significant differences in K content across the various growth stages except for K content in the leaves between 15 and 18 year olds, and between 21 and 25 year olds, the

Table 1: Nutrient content of Chinese fir plantations at different growth stages (g/kg)

Growth stage	Nutrient element	Trunk	Bark	Twig	Needle	Root
7-year old	N	0.92±0.066aA	3.58±0.1968bA	3.54±0.20cA	11.86±0.94dA	2.84±0.18eA
	P	0.12±0.016aM	0.37±0.028bM	0.40±0.03cM	1.09±0.07dM	0.33±0.03eM
	K	0.34±0.036aW	3.61±0.218bW	3.92±0.17cW	9.18±0.51dW	2.73±0.16eW
	Ca	0.63±0. 046aP	5.80±0.338bP	6.08±0.31cP	10.87±0.62dP	3.62±0.20eP
	Mg	0.68±0.416aN	0.72±0.388bN	1.75±0.09cN	2.78±0.14dN	1.05±0.07eN
	Sum	2.96a	14.18b	15.72c	33.07d	10.57e
12-year old	N	1.19±0.076aA	4.12±0.238bA	3.97±0.21cA	15.92±0.19dA	3.19±0.17eA
	P	0.18±0. 016aM	0.44±0.038bM	0.62±0.04cM	1.22±0.07dM	0.43±0.02eM
	K	0.37±0.036aW	3.67±0.208bW	4.16±0.23cW	9.63±0.51dW	2.94±0.16eW
	Ca	0.66±0.046aP	6.80±0.398bP	6.84±0.39cP	11.79±0.61dP	3.76±0.21eP
	Mg	0.78±0.046aN	0.98±0.068bN	2.21±0.13cN	2.89±0.15dN	1.28±0.07eN
	Sum	3.16 a	15.91 b	17.90 c	41.40 d	11.50 e
15-year old	N	1.10±0.586aA	4.01±0.238bA	3.75±0.20cA	15.40±0.18dA	3.10±0.17eA
	P	0.15±0.016aM	0.41±0028bM	0.59±0.03cM	1.18±0.07dM	0.41±0.03eM
	K	0.32±0.026aW	3.53±0.218bW	3.89±0.20cW	9.27±0.51dW	2.84±0.17eW
	Ca	0.64±0.046aP	6.92±0.368bP	6.80±0.38cP	11.81±0.59dP	3.79±0.20eP
	Mg	0.70±0.046aN	0.92±0.058bN	2.09±0.12cN	2.86±0.19dN	1.20±0.64eN
	Sum	2.91 a	15.79 b	17.12 c	40.52 d	11.34 e
18-year old	N	1.05±0.546aA	3.98±0.388bA	3.44±0.17cA	14.89±0.17dA	2.95±0.17eA
	P	0.13±0.016aM	0.40±0.028bM	0.57±0.03cM	1.17±0.07dM	0.40±0.02eM
	K	0.34±0.026aW	3.54±0.208bW	3.92±0.36cW	9.29±0.47dW	2.85±0.16eW
	Ca	0.65±0.046aP	6.93±0.358bP	6.81±0.38cP	11.88±0.54dP	3.81±0.21eP
	Mg	0.71±0.046aN	0.91±0.058bN	1.98±0.10cN	2.85±0.17dN	1.22±0.14eN
	Sum	2.84 a	15.76 b	16.72 c	40.08 d	11.23 e
21-year old	N	0.98±0.066aA	3.88±0.218bA	3.26±0.20cA	13.94±0.74dA	2.89±0.16eA
	P	0.12±0.016aM	0.41±0.028bM	0.58±0.03cM	1.18±0.07dM	0.41±0.03eM
	K	0.35±0.026aW	3.55±0.198bW	3.93±0.20cW	9.31±0.50dW	2.86±0.16eW
	Ca	0.68±0. 046aP	6.97±0.388bP	6.83±0.38cP	11.90±0.59dP	3.85±0.20eP
	Mg	0.69±0.046aN	0.92±0.068bN	1.97±0.10cN	2.87±0.16dN	1.19±0.07eN
	Sum	2.82 a	15.73 b	16.57 c	39.20 d	11.20 e
25-year old	N	0.90±0.056aA	3.81±0.218bA	3.28±0.22cA	14.11±0.79dA	2.85±0.17eA
	P	0.12±0.016aM	0.41±0.238bM	0.57±0.04cM	1.17±0.06dM	0.41±0.02eM
	K	0.37±0.026aW	3.57±0.208bW	3.95±0.19cW	9.34±0.45dW	2.89±0.15eW
	Ca	0.70±0.036aP	6.99±0.348bP	6.86±0.38cP	11.93±0.58dP	3.89±0.09eP
	Mg	0.70±0.036aN	0.91±0.068bN	1.98±0.09cN	2.86±0.16dN	1.20±0.07eN
	Sum	2.79 a	15.69 b	16.66 c	39.41 d	11.24 e

Different lowercase letters within rows indicate significant difference amongst the five organs of in terms of each nutrient element (p<0.05). Different capital letters within columns indicate significant differences amongst the five elements in the same organ (p<0.05)

twigs between 18, 21, and 25 year olds, and K content in the roots between 12, 21 and 25 year olds.

Nutrient content in the litter and dead roots

The nutrient content was higher in the litter than that in the dead roots at the same growth stages, and the content of each element significantly (P<0.05) differed between the litter and dead roots (Table 3). The relationship between nutrient content whether in the litter or dead roots and growth stages showed the same tendency as the tree layer.

The N, K, and Ca content of the litter and dead roots significantly differed across the various growth stages (P<0. 05) (Table 3). The P content of the litter also significantly differed across the growth stages, but not in the dead roots. The variation of Mg content with each growth stage was more complex. There were no significant differences in the Mg content of the litter amongst the growth stages except for at 12-years. In contrast, the dead roots differed across all growth stages except for between 12 and 15 years, and 21 and 25 years.

Discussion

The nutrient content of the fir organs varied with growth stage in the Suining forest, Hunan Province, resulting in significant differences in nutrient content in same fir organs at different growth stages. Regarding nutrient content in the different organs at the same growth stages, the trends were determined by the genetic and physiological characteristics of the Chinese fir, which was similar for Taoyuan (Feng *et al.* 1985) in the Hunan Province. The order and value of the various nutrients in the same organs at the same growth stages showed some differences. Taking the nutrient content in the needles at the 12-year old fir forest stage as an example, the content of N, Ca, K, Mg and P were 15.92, 11.97, 9.63, 2.94 and 1.22 g/kg in Suining, and 15.22, 11.60, 9.80, 2.80, 0.90 g/kg in Huitong, respectively, showing the same trend as N>Ca>K>Mg>P. However, the order of the above elements represented N>K>Ca>Mg>P, for which the contents were 10.22, 79.13, 5.10, 2.42 and 0.62 g/kg, respectively. Although the three sample areas were all located in the mid-subtropical zone, the microclimate conditions of Zhuting and the other two areas (which were

Table 2: Nutrient content in the undergrowth vegetation of Chinese Fir Plantations at different growth stages (g/kg)

Growth stage	Leaf					Twig (including small stem)					Root				
	N	P	K	Ca	Mg	N	P	K	Ca	Mg	N	P	K	Ca	Mg
7-year old	13.27aA	1.53bA	11.27cA	8.14dA	2.24eA	4.21fA	0.52mA	5.21nA	4.12hA	1.18kA	3.68tA	0.50yA	2.70wA	3.31sA	0.82rA
12-year old	15.87aB	159bB	11.96cB	8.25dB	2.67eB	4.55fB	0.64mB	5.99nB	4.24hB	1.59kB	3.91tB	0.60yB	2.54wD	3.47sB	1.09rB
15-year old	15.72aC	1.58bB	11.03cC	8.37dC	2.59eC	4.42fC	0.62mB	6.04nC	4.59hC	1.52kC	3.82tC	0.58yB	2.83wC	3.64sC	1.02rC
18-year old	15.44aD	1.57bB	10.99cC	8.43cD	2.54eD	4.35fD	0.60mB	5.87nD	4.62hD	1.47kD	3.73tD	0.57yB	2.66wE	3.72sD	0.91rD
21-year old	15.36aE	1.58bB	10.84cD	8.48dE	2.48eE	4.29fE	0.60mB	5.85nD	4.69hE	1.41kE	3.64tE	0.55yB	2.57wD	3.82sE	0.84rE
25-year old	15.22aF	1.56bB	10.81cD	5.53dF	2.43eF	4.22fF	0.59mB	5.82nD	4.74hF	1.34kF	3.50tF	0.55yB	2.53wD	3.92sF	0.75rF

Different lowercase letters within rows indicate significant differences ($p<0.05$). Different capital letters within columns indicate significant differences ($p<0.05$)

Table 3: Nutrient content of the litter and dead roots of Chinese Fir Plantation at different growth stages (g/kg)

Growth	Litter					Dead root				
stage	N	P	K	Ca	Mg	N	P	K	Ca	Mg
7 years old	5.80±0.23aA	0.86±0.05bA	0.55±0.03cA	9.98±0.58dA	1.28±0.77eA	2.42±0.16fA	0.32±0.02mA	0.40±0.03nA	3.32±0.17hA	0.77±0.05kA
12 years old	6.29±0.38aB	0.93±0.05bB	0.64±0.04cB	10.37±0.59dB	1.35±0.73eB	2.58±0.17fB	0.34±0.02mA	0.57±0.03nB	3.44±0.18hB	1.01±0.06kB
15 years old	6.12±0.32aC	0.87±0.05bC	0.58±0.03cC	10.59±0.61dC	1.27±0.68eA	2.53±0.15fC	0.32±0.02mA	0.50±0.02nC	3.52±0.17hC	0.99±0.05kB
18 years old	5.94±0.29aD	0.76±0.04bD	0.49±0.03cD	11.07±0.57dD	1.28±0.65eA	2..47±0.16fD	0.33±0.02mA	0.43±0.03nD	3.61±0.18hD	0.96±0.05kC
21 years old	5.82±0.29aE	0.71±0.05bE	0.43±0.02cE	11.65±0.63dE	1.29±0.59eA	2.42±0.14fE	0.34±0.02mA	0.38±0.02nE	3.75±0.20hE	0.91±0.06kD
25 years old	5.75±0.30aF	0.69±0.04bF	0.36±0.03cF	11.87±0.66dF	1.30±0.67eA	2.36±0.13fF	0.33±0.02mA	0.32±0.02nF	3.84±0.21hF	0.93±0.05kD

Different lowercase letters within rows indicate significant differences ($p<0.05$). Different capital letters within columns indicate significant differences ($p<0.05$)

adjacent) still differed. The soil thickness and N, P and K content were lower in Zhuting than that of Suining and Huitong (Feng *et al.* 1985). Differences in the micro-environmental conditions and soil fertility levels could lead to these differences in the plant body (Chen *et al.* 1999).

Firstly, the nutrient content of the different forest trees differed. For example, the order of nutrient content in the same organs at each growth stage of Chinese fir were needles> twigs > bark > roots > trunk, compared to needle > bark > twigs > roots > trunk of *A. mangium* (He *et al.* 2007) and *P. tabulaeformis* (Shen *et al.* 1985). The nutrient content in the plant organs of Chinese fir were related to organ function. Needles were the main photosynthesis organ that provided the biomass. Fir had a shorter growth period and required higher levels of nutrients for its growth and metabolism, and as such, a greater nutrient content were required compared to other organs. Physiological and biochemical activities were weakest in the plant trunk, and as such, nutrients were easily consumed or transferred, resulting in a lower nutrient content. Plant twigs and barks were the transport organs, and specific nutrients were required to satisfy their functions, leading to a higher nutrient content. In addition, plant twigs and bark had supporting functions, so that Ca content was higher than other nutrient elements. The roots were responsible for the absorption of nutrients and water, and supported the plant body. As such, the Ca content was higher than other nutrients. In addition, the roots were located closer to soil water and were protected and fed by adjacent soils, requiring lower levels of nutrients, resulting in a lower nutrient content. An array of factors led to changes in nutrient content in the plant tissues during the growth stage. Firstly, the levels of nutrients in plant tissues of different growth stages were determined by the genetic and physiological characteristics of the plants. The nutrient requirements for the production of 1 ton dry biomass declined at increasing growth stages when the Chinese fir entered the trunk wood stage (Tian and Xiang 2002), meaning the nutrient content in the plant tissue decreased with the development of the growth stages. Secondly, ambient climate factors also influenced the nutrient content of plant tissue. There were obvious variations in the precipitation and temperature across the years, which induced changes in the nutrient content of the plant tissue (Nie 1991). Finally, the organic components of plant tissue may have altered across the growth stages, for example, increases were observed in the degree of signification of the plant tissue. This may represent an important reason for the increase in Ca content in Chinese fir organs at increasing growth stages.

Secondly, the order of the various nutrient elements in the same organ at near growth stages showed differences. For example, the order of various elements in the needles, twigs, bark and roots for 14-year-old *P. massonana* were K>N>Ca>Mg>P, and Ca>K>N>Mg>P in the trunk (Tian *et al.* 2002), compared to Ca>N>K>Mg>P in the bark and roots, N>Mg>Ca>K>P in the trunk, Ca>K>N>Mg>P in the twigs, and N>Ca>K>Mg>P in the needles of 14-year-old Chinese fir. The nutrient content in the undergrowth tissue was similar to that of the tree layer, which significantly increased from 7 to 12-years, and then gradually decreased with increasing growth stages at the trunk wood stage, except for Ca, which increased across the entire experiment period. That may be because the trees were small prior to age 7, with small canopy densities and adequate light under the forest. Heliophilous plants therefore dominated the undergrowth vegetation. Chinese fir from 7 to 12-years with lush cover limited the light in the understory, resulting in sciophilous plants being dominant. When the fir entered the trunk wood stage, the trees were self-pruning, and then improved the light conditions, so that the undergrowth vegetation was gradually replaced by heliophilous plants.

Studies have shown that heliophilous shrubs possess a higher caloric value and carbon content than sciophilous shrubs (Wang and Sun 2008). More information is required to assess whether sciophilous shrubs have a higher nutrient content than heliophilous shrubs.

Thirdly, there were extreme differences in nutrient content across the different tree species in the same organs according to growth stage. For example, the nutrient content in the trunk, bark, twigs, needles and roots of 7-year old *A. mangium* were 4.29, 22.22, 13.39, 40.96 and 7.12 g/kg (He *et al.* 2007), while those in the corresponding organs were 2.96, 14.18, 15.72, 33.07 and 10.57 g/kg in 7-year old Chinese fir. As a further example, the nutrient content in these organs were 23.50, 61.42, 57.93, 256.24 and 41.04 g/kg in 25-year old *P. tabulaeformis* (Shen *et al.* 1985), compared to 2.79, 15.96, 16.66, 39.41 and 11.24 g/kg, respectively, in 25-year old Chinese fir. Regarding differences in the genetic and physiological characteristics of the different tree species, there were differences in site conditions. The different site conditions made a contribution to the differences in the nutrient content of the plant body.

Finally, consistent with previous studies, the undergrowth vegetation contained a higher nutrient content than that of the tree layer. This was mediated by the spatial distribution patterns of the plants (living) as the nutrient content of the forest ecosystem generally increased from the upper layer to the lower layer, namely, the tree layer showed the lowest nutrient content, followed by the bush layer, whilst the grass layer showed the highest nutrient content (Zhang *et al*. 2006).

As for the litter content, the nutrient content was lower than that of living plants, which was determined by the nutrient biological cycle in the plant body. That was probably due to the fir and undergrowth vegetation being the source of the litter and dead roots, and as such, the nutrient content in the former determined the nutrient content in the latter. Furthermore, the nutrient content of needles, twigs and bark were higher than that in the roots of Chinese fir based on previous analysis, which led to the nutrient content in the litter exceeding that of dead roots. This study showed that the nutrient content of the litter was lower than that of corresponding living plants (whether in the tree layers or undergrowth vegetation). This was because some of the nutrients in the aging tissue had transferred to other living sites prior to falling.

Conclusion

Nutrient content in the same organs and site conditions differed amongst the different growth stages of Chinese fir, suggesting that some deviation exists when only a single growth stage is used to demonstrate the relationship between the biomass, nutrient content and nutrient utilization, and the accumulation and circulation of the entire growth process. We highlight the consistency of the spatial scale and the continuity of the time scale, which could overcome the heterogeneity of various site conditions. The defects in "space for time" methods could therefore be circumvented producing data of more physiological relevance. These findings provide improved guidance for production practices.

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Author Contributions

Yun-ye Deng and Xian-jun Yang carried out the concepts, design, definition of intellectual content, literature search, data acquisition, data analysis and manuscript preparation. Xiao-yi Xing and Ying-hui Li provided assistance for data acquisition, data analysis and statistical analysis. Li-xia He and Fei Ni performed manuscript review. All authors have read and approved the content of the manuscript.

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