

## **Response of Earthworms' Ecological Groups to Decay Degree of Dead Trees (Case study: Sardabrood Forest of Chalous, Iran)**

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### **ABSTRACT**

*Dead tree is an important ecological factor for changes and heterogeneity creation of soil characters that attracts different livings. Due to investigation the decay degree effects of downed tree on soil characters and earthworms ecological groups, the Sardabrood forest of Chalous studied that is located in Mazandaran province, northern Iran. For this purpose, 306.2 ha areas considered at 700 - 1300 m altitude range and twenty seven downed trees of beech and hornbeam species were found. Decay degree of downed tree classified in four classes (DC1, DC2, DC3 and DC4). Whole of dead trees selected as the center of sample plots and mixed soil samples were taken from theirs besides and along at 0 -10, 10 - 20 and 20 - 30cm depth. Soil acidity, water content, total carbon, total nitrogen and carbon to nitrogen ratio measured in the laboratory. The earthworms were collected simultaneously with the soil sampling by hand sorting. The maximum of acidity and water content considered in DC4 of dead trees and significant differences were found. But, soil depth had no significant differences for these characters. Also, the most amounts of soil carbon and nitrogen found in DC4 of dead trees and upper layers of soil. DC1 of dead trees and soil lower depths devoted the maximum of carbon to nitrogen ratio. Principal component analysis (PCA) showed that the most assemblage of earthworms were around of dead trees with high decay degree (DC4). Number and biomass of earthworms group in dead trees positions indicating endogeics are due to creating significant statistical differences between beech and hornbeam downed trees.*

**Key words:** Decay degree, soil character, beech and hornbeam, PCA.

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### **INTRODUCTION**

In the Caspian region, with a forest surface area of about 1500000 ha, oriental beech (*Fagus orientalis* Lipsky) is an important climax species, together with species such as Hornbeam (*Carpinus betulus* L.), Caucasian oak (*Quercus castaneifolia* C.A.M.), Caucasian zelkova (*Zelkova crenate* Desf.), Persian iron wood (*Parrotia persica* DC. (C.A.M.) and Caspian honey – locust (*Gleditschia caspica* Desf). The beech is a main species in this mixed broadleaved forests and appears stands wise in the mountainous sites of northern slopes of Elburz Mountains (700 - 2200 m.a.s.l.). In the north of Iran, pure and mixed oriental beech forests cover 17.6 per cent of the standing volume. Beech is the most valuable wood - producing species in the Caspian forests. Old beech trees can grow taller than 40 m and exceed a diameter at breast height larger than 1.5 m [35, 44]. In unmanaged forest ecosystems, the main causes of small - scale soil heterogeneity are micro - relief, soil parent material, spatial distribution of trees and tree species and uprooting of trees and dead trees creation [10]. Dead wood is an important component in natural forests. It is widely regarded as an important aspect of forest biodiversity forming key habitats for many species. For example invertebrates, fungi, bryophytes, lichens, birds and mammals depend on or utilize dead woods as source of food or shelter [42]. Dead wood is also an important long term nutrient storage [16], the carbon content adds significantly to the overall carbon storage of forest ecosystems [12, 50], and humification process secures a continuous supply of organic material to the soil [38, 51, 52].

Dead wood is not an optional extra, but a critical component in forest functioning, which plays five major roles in the ecology of healthy, natural forest including: maintaining forest productivity by providing organic matter, moisture, nutrients and regeneration sites for conifers - some tree species germinate preferentially on logs; providing habitats for creatures that live, feed or nest in cavities in dead and dying timber, and for aquatic creatures that live in pools created by fallen logs and branches; supplying a food source for specialized feeders such as beetles and for fungi and bacteria; stabilizing the forest by helping to preserve slope and surface stability and preventing soil erosion in the event of storms, heavy rainfall and other climatic extremes; storing carbon in the long term, thus mitigating some of the impacts of climatic change [43]. Dead wood isn't a single habitat, but instead a complex range of different microhabitats, which change and evolve over time. The quality of deadwood, and its usefulness for different species, depends on how long it has been decaying and also on the tree species, age at time of death, cause of death, position (standing, fallen, etc) and size, and on the surrounding climatic conditions [30]. Earthworms are perhaps the most important soil organisms in terms of their influence on organic matter breakdown, soil structural development, and nutrient cycling, especially in productive ecosystems [18]. Aristotle called them the "instincts of the earth" and the eminent nineteenth century biologist, Charles Darwin, aspect many years observing their major influence on the formation of humus and transport of soil [17]. Despite of the vast increase in scientific literature on earthworms in recent years, much remains to be known in their basic biology and ecology [18, 28]. It is not clear whether earthworm populations are mainly controlled by the amount of food, its quality, or the chemical properties of their environment [3, 11, 41]. Therefore, determining the relation among biomass and diversity of earthworms with downed tree disturbances (dead trees) and edaphic conditions are essential for management of forest ecosystems. The main aim of the present study was to investigate of downed tree effects on soil properties, earthworm biomass and species diversity in mountainous forests of Iran that is the first survey in these forests and no data have yet been published.

## MATERIALS AND METHODS

**Study area:** This research performed in Sardabrood forests that are located in the lowland and midland of Mazandaran province in north of Iran with the area of 2347 ha. (Between 36° 37' 30", 36° 40' 52" northern latitude, and between 51° 7' 50", 51° 12' 51" eastern longitude). The maximum elevation is 1400m and minimum is 50m. Minimum temperature in December (7.5°C) and the highest temperature in June (24.6°C) are recorded, respectively. Mean annual precipitation of the study area were from 237.6 to 47.5 mm at the Noushahr city metrological station, which is 10Km far from the study area. The soils are deep, moderately well drained. They have textures of silty clay and clay loam with pH of 4.9 to 6.3. Bedrock is sandstone with silting and argillite, and lime stone. Presence of logged and bare roots of trees is indicating rooting restrictions and soil heavy texture [1].

**Soil sampling and analysis:** Due to this survey, twenty seven downed tree in mixed beech forests were found at 306.2 ha areas. Seventeen trees dominated by beech (*Fagus orientalis* Lipsky) and ten by hornbeam (*Carpinus betulus* L.) at 700 - 1300 m altitude range (Table 1). Decay degree of downed trees classified in four classes including (Fig. 1): the tree is dead recently, cambium still green, crown intact (DC1); bark sloughing, usually fine longitudinal shakes in the wood, twigs sloping (DC2); the decay is advanced, spreading of the longitudinal shakes to furrowes (DC3); the wood is decayed completely, log collapsing, wood friable, crown completely decomposed (DC4) [23].

Whole of dead trees selected as the center of sample plots and mixed soil samples excavated from theirs besides and along. Soil acidity, water content, total carbon, total nitrogen and carbon to nitrogen ratio measured in the laboratory. Soil samples were taken at 0 - 10, 10 - 20 and 20 - 30cm depth from all sites. Large live plant material (root and shoots) and pebbles in each sample were separated by hand and discarded. The soil samples were air - dried and sieved. Soil acidity (with an electrode), water content (by drying soil samples at 105° C for 24 hours), total carbon (Walkey and Black method), total nitrogen (Kjeldahl method) and carbon to nitrogen ratio measured in the laboratory [40].

**Table 1. Downed trees characteristics of beech and hornbeam**

Species	Tree number	Average of D. B. H. (cm)	Average of altitude (m)	Dominant slope	Slope aspect
<i>Fagus orientalis</i> Lipsky	17	45.35 (35 - 52)	1202.1 (1110 - 1295)	40 - 50	Northeast
<i>Carpinus betulus</i> L.	10	48.60 (42 - 52)	771.5 (725 - 910)	40 - 50	Northeast
Beech site included number of 3, 4, 6, and 4 downed trees in DC1, DC2, DC3 and DC4, respectively. Hornbeam site included number of 3, 4 and 3 downed trees in DC1, DC2 and DC4, respectively (DC3 wasn't considered in hornbeam site).					

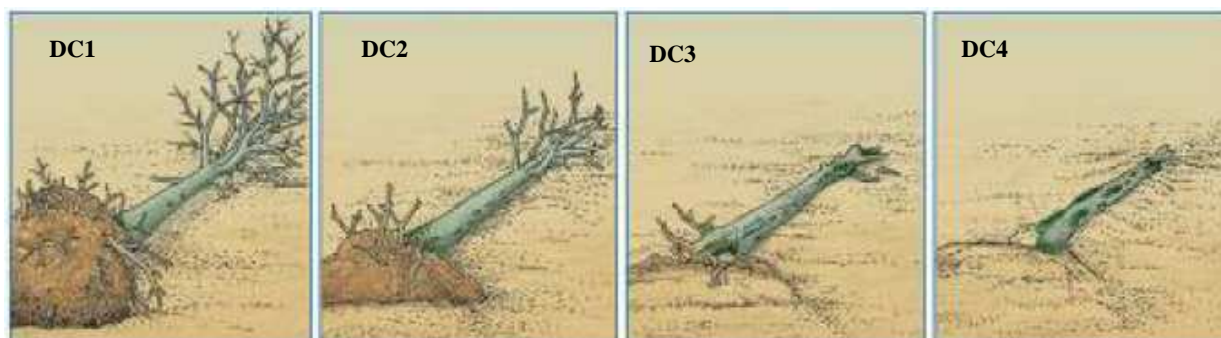


Fig. 1. Decomposition key for woody debris of beech and hornbeam

**Identification and differentiation of earthworms:** The earthworms were collected simultaneously with the soil sampling by hand sorting, washed in water and weighed with mili gram precision. Species of earthworms were identified (epigeic, anecic, and endogeic) by external characteristics using the key of BOUCH [8]. **Epigeic** worms feed on plant litter, dwell on the soil surface or within the litter layer, tend to be heavily pigmented, and are small to medium sized. **Anecic** worms feed on plant litter and soil, live in nearly vertical permanent burrows, are dorsally pigmented, and large. **Endogeic** species are soil - feeders, are not heavily pigmented, from extensive horizontal burrow systems, and range in size from small to large. Earthworm species do not always fall clearly into these three main categories and may even exhibit traits of different groups at different life stages or under different environmental conditions [2, 5, 7, 9, 13, 14, 21, 22, 29, 31, 32, 34, 36, 39, 45, 47, 48]. Biomass was defined as the weight of the worms after drying for 48 hours on filter paper at room temperature (60°C) [8].



Fig. 2. Earthworm's representative of different ecological groups. (a) Epigeic, (b) Endogeic, (c) Anecic.

**Data analysis:** Kolomogorov - Smirnov test used as normality test and Levene test for data homogeneity test. Analysis of variance (one - way ANOVA) and Duncan comparison were used to find differences in soil characteristics, earthworm's number and biomass of the decay different classes. Square - root method used for normality of data, because in some cases there was no homogeneity of variance. Analysis of whole data was done in SPSS Ver. 13.5 of statistical program. Factor analysis is statistic technique for achievement to complex relationships among variables. For this purpose, relationships among decay classes, soil characteristics and earthworm groups were analyzed by Principle Component Analysis [25].

## RESULTS AND DISCUSSION

**Soil characteristics:** Analysis of variance is indicating soil characteristics have significant differences in decay different degrees of beech and hornbeam downed trees (Table 2). The maximum of acidity were observed in DC4 and this factor had significant differences with the other decay classes, but soil depth had no significant differences (Table 2). The most water content (moisture) devoted in DC4 and soil depth had no significant differences also (Table 2). The most amounts of carbon and nitrogen related to DC4 and soil upper layers, but DC1 and soil lower layers had the most value of carbon to nitrogen ratio (Table 2).

The studied soils were found acidic and alkaline in around of beech and hornbeam dead trees, respectively. The most amount of pH was considered in DC4 of hornbeam downed tree and DC1 to DC3 of beech downed trees. Totally, faguetum litters have low pH and carpinetum litters have alkaline conditions that are due to acidic and alkaline of soil [27]. The results of this research showed that the alkaline condition of soil increased along time and with more decay of hornbeam dead trees, that is probably for the presence released different compounds because of more decay and decomposition of dead trees by destructive factors (fungi, insects and invertebrates). Whereas beech dead trees had a different condition that is by reason of edaphic factors, elements, components and beech inner structure. Performed survey showed no regular changes among soil layers related to acidity amounts. This subject is linked to irregular

changes of lime in lower layers and humus, organic matter presence in soil surface [4]. Water content increased significantly with increasing of dead trees decay degree. Motta [26] reported that dead tree presence will create more suitable conditions for increasing water uptake in soil of forest ecosystems. Thus, with increasing of dead tree decay degree, their contents and components will create more sponge condition that is due to capability increase of moisture uptake and maintenance by soil. Gap opening in canopy closure (by reason of tree fall) is due to more intense contact of sunlight to forest floor, moisture more evaporation of soil surface and elevating the water content to soil surface [6]. In current study, moisture percent is reduced in surface soils by reason straight shine of sun light, but the soil lower layers had more amounts of water contents.

**Table 2. Mean of soil characteristics in decay different degrees of downed trees**

Downed tree species / Soil characteristics			pH	Water content	Carbon	Nitrogen	C/N ratio
Beech	Decay Class	1	6.51 (0.00)a	31.42 (0.22)d	2.81 (0.08)c	0.15 (0.00)c	17.92 (0.33)a
		2	6.45 (0.01)a	35.70 (0.23)c	2.97 (0.04)b	0.18 (0.00)b	16.70 (0.61)b
		3	6.43 (0.01)a	38.58 (0.22)b	2.98 (0.04)b	0.22 (0.00)a	13.51 (0.17)c
		4	6.15 (0.04)b	43.78 (0.29)a	3.13 (0.01)a	0.24 (0.00)a	13.15 (0.42)c
	Soil depth (cm)	0 - 10	6.43 (0.01)	36.94 (1.00)	3.15 (0.01)a	0.23 (0.00)a	13.72 (0.45)b
		10 - 20	6.34 (0.04)	37.95 (1.04)	3.00 (0.02)b	0.20 (0.00)b	14.73 (0.50)b
		20 - 30	6.39 (0.04)	38.69 (1.05)	2.80 (0.05)c	0.17 (0.00)c	16.41 (0.59)a
Hornbeam	Decay Class	1	7.61 (0.00)b	29.53 (0.19)c	2.77 (0.07)c	0.18 (0.01)b	15.08 (0.71)a
		2	7.54 (0.03)b	34.57 (0.17)b	2.96 (0.02)b	0.21 (0.01)b	14.47 (0.83)a
		4	8.07 (0.02)a	41.48 (0.33)a	3.14 (0.02)a	0.26 (0.00)a	11.92 (0.35)b
		0 - 10	7.73 (0.66)	34.38 (1.50)	3.07 (0.03)a	0.25 (0.01)a	12.45 (0.56)b
	Soil depth (cm)	10 - 20	7.73 (0.09)	35.37 (1.59)	2.97 (0.04)ab	0.21 (0.01)ab	14.10 (0.58)ab
		20 - 30	7.71 (0.08)	35.65 (1.57)	2.83 (0.07)b	0.19 (0.01)b	15.11 (1.02)a

Values are the means  $\pm$ St. error of the mean (in parenthesis).

Within the same column the means followed by different letters are statistically different ( $P < 0.05$ ).

**Table 3. Mean of number and biomass of earthworms in decay different degrees of downed tree**

Downed tree species / Ecological groups		Number			Biomass				
		Epigeic	Anecic	Endogeic	Epigeic	Anecic	Endogeic		
Beech	Decay Class	1	0.79 (0.39)	2.08 (0.23)	2.16 (0.35)c	6.81 (3.40)	17.04 (1.70)	17.17 (3.35)c	
		2	1.88 (0.81)	2.4 (0.52)	3.10 (0.21)b	18.32 (7.85)	21.08 (4.56)	27.11 (2.77)b	
		3	5.52 (0.87)	2.47 (0.43)	3.17 (0.17)b	23.89 (8.25)	23.47 (4.14)	30.18 (1.96)b	
		4	3.16 (0.35)	2.94 (0.67)	4.42 (0.36)a	29.66 (9.52)	28.44 (6.62)	43.46 (3.76)a	
	Soil depth (cm)	0 - 10	6.66 (0.60)a	2.95 (0.21)b	2.40 (0.13)c	6277 (5.85)a	27.31 (2.06)b	20.93 (1.57)c	
		10 - 20	0.00 (0.00)b	4.07 (0.24)a	2.94 (0.21)b	0.00 (0.00)b	37.91 (2.75)a	26.77 (5.52)b	
		20 - 30	0.00 (0.00)b	0.46 (0.24)c	4.46 (0.22)a	0.00 (0.00)b	3.61 (1.95)c	43.14 (2.51)a	
	Hornbeam	Decay Class	1	1.51 (0.75)	2.68 (0.36)	3.37 (0.42)b	13.53 (6.76)	23.69 (2.95)	28.28 (4.03)b
			2	2.29 (0.98)	2.97 (0.70)	4.18 (0.25)ab	21.69 (9.28)	28.49 (6.75)	37.94 (3.21)ab
4			3.38 (0.69)	3.44 (0.90)	5.18 (0.58)a	32.19 (16.11)	28.23 (8.51)	50.95 (6.24)a	
Soil depth (cm)		0 - 10	7.16 (0.73)a	3.28 (0.27)b	3.15 (0.16)b	67.19 (7.34)a	31.20 (2.75)b	27.46 (1.53)b	
		10 - 20	0.00 (0.00)b	5.26 (0.33)a	3.72 (0.36)b	0.00 (0.00)b	48.7 (3.83)a	33.63 (4.29)b	
		20 - 30	0.00 (0.00)b	0.52 (0.28)c	5.76 (0.33)a	0.00 (0.00)b	5.03 (2.70)c	55.76 (3.90)a	

Values are the means  $\pm$ St. error of the mean (in parenthesis).

Within the same column the means followed by different letters are statistically different ( $P < 0.05$ ).

The highest value of soil carbon and nitrogen found in DC4 of dead trees. Vitousek and Denslow [46] demonstrated that downed trees are an effective factor for increasing the amounts of carbon and nitrogen in forest soil. Totally, dead tree and decayed trees will storage the carbon and nitrogen in long term and release these elements to forest soil gradual by decomposition process [15]. In our research, also with increasing of decay and decomposition degree in long term, more amounts of carbon and nitrogen were released in studied soil. On the other hand, the maximum of carbon and nitrogen considered in soil upper layers. Kooch, et al. [20] in his research showed that the carbon amount will reduce with increasing soil depth that is related to litter and plant residues accumulate in soil surface. Also, he reported that carbon character has direct relation with nitrogen amount in soil surface as the soil carbon and nitrogen percent will decrease with increasing of soil depth. These relations were found in our study also. Carbon to nitrogen



ratio of soil is depended to carbon and nitrogen percent in forest ecosystem soil. Thus, the obtained results showed that the most value C/N ratio devoted in DC1 and this ratio will decreased in long term with increasing of dead tree decomposition. The most amount carbon to nitrogen ratio was considered in lower layers of soil linked to carbon and nitrogen percent.

**Ecological groups of earthworms:** Analysis of data showed that number and biomass of endogeic ecological groups had significant differences among decay different degrees of downed trees (Table 3). Earthworm's number and biomass of endogeic had more amounts in DC4 and the other ecological groups of earthworms (epigeic and anecic) had no significant differences (Table 3). The most earthworms number and biomass of epigeic, anecic and endogeic were found in 0 - 10, 10 - 20 and 20 - 30cm soil depths, respectively. Principle component analysis (PCA) is indicating the most assemblage of earthworm ecological groups is around of downed trees with high decay degrees (DC4) (Fig. 3 and 4). Also, investigation of earthworms groups in location of beech and hornbeam downed trees showed that endogeic number and biomass had significant statistical differences between mentioned downed trees (Table 4).

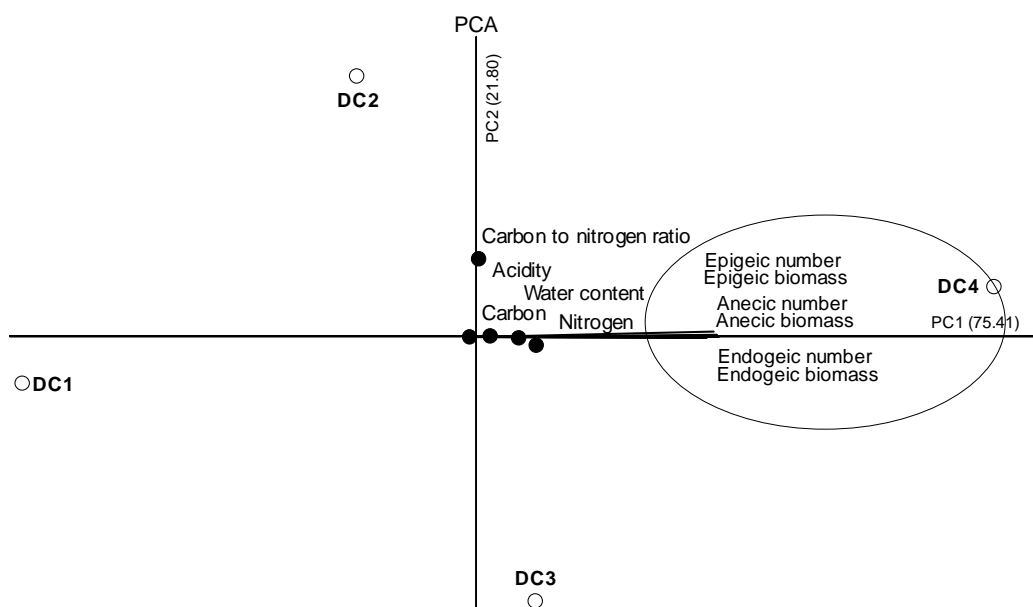


Fig. 3. PCA biplots of decay class, soil characteristics and earthworm group in beech downed tree location

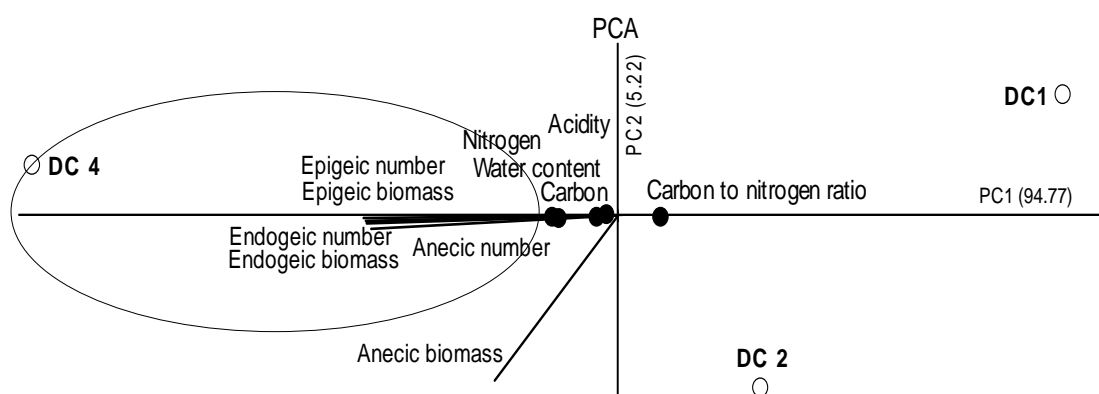


Fig. 4. PCA biplots of decay class, soil characteristics and earthworm group in hornbeam downed tree location

Table 4. Mean of earthworm's number and biomass in beech and hornbeam downed trees

Ecological groups of earthworm	Number						Biomass					
	Epigeic		Anecic		Endogeic		Epigeic		Anecic		Endogeic	
Downed tree species	B	H	B	H	B	H	B	H	B	H	B	H
Mean	2.05	2.38	2.51	3.02	3.32	4.21	19.30	22.39	22.65	28.23	30.34	38.95
Sig.	0.71 <sup>ns</sup>		0.31 <sup>ns</sup>		0.01**		0.71 <sup>ns</sup>		0.24 <sup>ns</sup>		0.03*	

(B): Beech. (H): Hornbeam. \*\* Different is significant at the 0.01 level.

\*Different is significant at the 0.05 level. (ns): Non significant differences ( $P > 0.05$ ).

Totally, the most earthworms are sensitive to soil acidity as their number and biomass reduce in soils with low pH. On basis of carried out researches, earthworms prefer pH inclined to buffer [18]. Thus, total comparison of beech and hornbeam dead trees is indicating more presence and biomass of earthworms in hornbeam dead trees that devoted the more amounts of soil acidity (alkaline) in compare to beech dead trees. The results of our study showed that DC4 of downed trees had the most density and biomass of earthworms (although, only number and biomass of endogeic ecological groups had significant differences). It is considerable that more assemblage of endogeic group can be related to water contents in lower layers of soil. Almost, 80 to 90 % of earthworm's fresh weight constituted of water, thus soil moisture are essential for their live and will kill by reason of soil drying [19]. Pay attention to, lower horizons of soil devoted in endogeic groups and to perceive these earthworms groups have high excavation ability [17] thus; they had more assemblage in comparison to the other groups of earthworms. On the other hand, Mboukou - Kimbatsa, et al. [24] demonstrated the effect of soil nitrogen on earthworm's density and biomass as positive. In current research, DC4 of dead trees had the most amount of soil nitrogen and included high density and biomass of earthworms that is according to Mboukou - Kimbatsa, et al. [24] description. But, the low C/N ratio of soil is the other effective factor that can be considered in relation to earthworms' groups' assemblage in DC4 of dead trees. Wood [49] in his research resulted that soil mineral matters are necessity for earthworms' growth and their biomass will increase in soil with low C/N ratio. Rahmani [33] resulted that earthworm population are effected by C/N ratio, as density and biomass of earthworms are reduced by increasing C/N ratio. Neyrinck, et al. [29] reported that the low C/N under canopy of Maple is due to more assemblage of earthworms. Antunes, et al. (2008) demonstrated soil C/N as an abiotic factor and the most important effective factor on density and biomass of earthworms in forest ecosystem. Kooch, et al. [19] introduced the soil C/N factor as the most effective factor on earthworm density and biomass in ecosystem units of Chalous lowland forests (North of Iran). In our study, the least amounts of soil C/N found in DC4 of dead trees, thus more earthworms' density and biomass considered in this decay class. Principal component analysis confirmed the accuracy of above phrase.

As figure 2 showed the C/N ratio and DC4 of dead trees are laid in two sides of main principle components that is indicating their vice versa relation each other. Also, this figure is showing assemblage of earthworm different groups in DC4 (of course, in hornbeam dead trees, anecic biomass is separated from the other earthworms groups that can be related to earthworm age and other uninvestigated factors in this survey). The most number and biomass of epigeic found in upper soil layer. As mentioned before, this earthworm group haven't high excavation ability, thus they will assemblage in superficial horizons that soils are finer (especially with presence the decay high degree of dead trees). The most earthworms (especially epigeic group) prefer rich environments of nutrient source [18]. The presence of dead wood can be as effective nutrient source for their presence. Anecic and endogeic species are more resistant to soil inappropriate textures and have high excavation ability in compare to epigeic [17]; therefore these earthworm groups were considered in soil lower layers. Totally, it is mentionable that downed trees or dead trees can create a new niche for many of livings and play a pivotal role in nutrient cycle [37].

## CONCLUSION

In this research tried to survey of dead trees role on earthworm density and biomass and is as the first research that analyzed about this subject in the north Mountainous forest of Iran. Understanding the ecological effects of dead trees on soil livings can present accuracy perceive of forest soil ecology to be applicable in managed forests. It is proposed that similar researches should be carried out with respect to dead trees effects on soil nutrients elements (especially N, P, K), and also the response of earthworm groups to these nutrients elements. Because of dead trees are rich sources of nutrient elements that have different ecological effects on forest ecosystem soil.

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