



IMPACT OF *CEDRUS LIBANI* AFFORESTATION ON SOIL CARBON AND NITROGEN STOCKS IN THE UPPER MEDITERRANEAN BASIN

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ABSTRACT: Soils, as the most dynamic and complex components of terrestrial ecosystems, serve as crucial sinks for atmospheric carbon storage and retention. Human interventions in soil management significantly influence the amount of carbon and nitrogen stored or sequestered. This study investigated the effects of afforestation using Cedrus libani A. Rich (Lebanese cedar or Taurus cedar) at two different ages (10 and 25 years) in the Upper Mediterranean basin on soil organic carbon and nitrogen stocks. The afforestation was conducted on previously bare lands for soil conservation purposes. A total of 45 soil samples were collected from topsoil (0-10 cm): 15 samples were randomly taken from two different times (2000 and 2015) afforested areas and 15 from non-afforested (control) land. Soil organic carbon (SOC), total nitrogen (TN), and bulk density (BD) analyses were performed on these samples. To calculate soil organic carbon stocks in tons per hectare, bulk density (BD) was estimated using the SOC and soil mass equation. The results revealed a substantial increase in carbon and nitrogen storage in the afforested areas, depending on tree age. Specifically, organic carbon and nitrogen stocks in the topsoil of 25-year-old and 10-year-old afforestation sites were 65% and 48% higher, respectively, than in control soils. Carbon and nitrogen storage followed the trend: 25-year > 10-year > 0-control. The highest total nitrogen content (0.78%) was observed in 10-year-old cedar afforestation sites. While BD values did not differ significantly among afforested areas, the control areas showed distinct differences from the afforested sites. This study demonstrates that age-protected cedar afforestation significantly enhances carbon and nitrogen sequestration in previously bare soils, highlighting its importance for soil conservation and ecosystem sustainability.

Keywords: Cedar, afforestation, Karst, carbon sequestration, carbon stocks, nitrogen

CEDRUS LIBANI AĞAÇLANDIRMASININ YUKARI AKDENİZ HAVZASINDA TOPRAK KARBON VE AZOT STOKLARI ÜZERINDEKİ ETKİSİ

ÖZET: Karasal ekosistemlerin en dinamik ve karmaşık yapıtaşı olan topraklar atmosferik karbonun tutularak depo edildiği en önemli yutakların başında gelmektedir. Topraklara yapılan müdahaleler depo edilen veya edilecek olan bu karbon ve azot miktarlarını değiştirmektedir. Bu çalışmada, toprak koruma amacıyla yukarı Akdeniz havzasındaki çıplak arazilerde 25 ve 10 yıllık Cedrus libani A. Rich (Lübnan sediri veya Toros sediri) kullanılarak yapılan ağaçlandırmaların toprakların organik karbon ve azot stoklarına olan etkileri araştırılmıştır. İki farklı zamanda (2000 ve 2015) ağaçlandırma yapılan alanlardan rastgele olarak 15 er adet ve ağaçlandırma yapılmayan çıplak bir araziden de 15 adet toprak alınmak suretiyle toplamda 45 adet toprak numunesi 0-10 cm derinliğinden alınmıştır. Toprak örneklerinde organik karbon (TOK), toplam azot (TA) ve hacim ağırlığı (HA) analizleri yapılmıştır. TOK ve TA stoklarını hektar başına ton cinsinden hesaplamak için HA, TOK veya TA ve toprak kütlesi eşitliğinden yararlanılarak tahmin edilmiştir. Elde edilen veriler sonucunda yaşa bağlı olarak çok ciddi miktarda sedir ağaçlandırma sahalarında karbon ve azot depolandığı, özellikle 25 ve 10 yaşlarındaki ağaçlandırma sahasındaki üst topraklarda depolanan karbon ve azot miktarının kontrol topraklarına nispeten sırasıyla %65 ve %48 daha fazla olduğu belirlenmiştir. Bu alanlardaki karbon ve azot depolama yaşa göre 25>10>0 şeklinde sıralanmıştır. Buna karşın toprakların toplam azot miktarlarında en yüksek değer 0.78 ile 10 yaşındaki sedir ağaçlandırma sahalarında bulunmuştur. İstatistiksel olarak ağaçlandırma yapılan alanlardaki hacim ağırlıkları birbirlerinden farklı olmadığı halde kontrol alanlarının hacim ağırlıkları ağaçlandırma yapılan alanlardan farklı bulunmuştur. Bu çalışma, yaşa bağlı olarak korunan sedir ağaçlandırma çalışmalarının çıplak topraklarındaki karbon ve azot miktarının önemli derecede arttırdığını göstermiştir.

Anahtar kelimeler: Sedir, ağaçlandırma, Karst, karbon tutulması, karbon stokları, azot

INTRODUCTION

Global warming is one of the most pressing environmental challenges affecting both our country and the world. To combat this issue, reducing atmospheric greenhouse gas emissions and enhancing carbon sinks are widely used strategies (Kokanç, 2014; Ozlu et al., 2022). Forests and forest soils serve as the primary terrestrial ecosystems capable of storing significant amounts of atmospheric carbon (Lee et al., 2016). In particular, substantial investments and efforts have been dedicated to afforestation, rehabilitation, and grazing management to protect soils in barren areas and enhance carbon sequestration potential. Through large-scale national initiatives such as afforestation campaigns, 'Türkiye Century,' and 'Breath to the Future,' extensive afforestation efforts have been undertaken across the country. Beyond their ecological and economic significance, these afforestation projects contribute to increased carbon storage in vegetation and soils, which is crucial for ecosystem sustainability. Therefore, understanding the dynamics of carbon sequestration in afforested and natural forest areas has become increasingly important (Kara et al., 2016; Babur et al., 2016; Lee et al, 2018).

Soils are among the most critical components of terrestrial ecosystems, capable of storing substantial amounts of organic carbon (Babur et al., 2021a; Dindaroğlu et al., 2021a). It is

estimated that global soil organic carbon (SOC) sequestration surpasses the atmospheric carbon pool by threefold and is four times greater than the biotic carbon pool (Lal, 2001). Recognizing this capacity, the Kyoto Protocol highlighted soil carbon storage as a key strategy for reducing CO₂ emissions, a major greenhouse gas (Ruiz-Sinoga et al., 2012). Furthermore, assessing and enhancing soil carbon storage was a central topic at the COP21 Climate Summit held in Paris in December 2015. In line with these efforts, the United Nations General Assembly adopted the 2030 Agenda for Sustainable Development, emphasizing the restoration of degraded soils (UNGA, 2015). Through conservation and development initiatives aligned with the Sustainable Development Goals (SDGs), the sustainable management of dynamic, limited, and fragile soil resources can be ensured (Jónsson et al., 2016). Given its significance, monitoring SOC stock dynamics has been proposed as a viable indicator for assessing land and soil changes within the SDG framework (Lorenz & Lal, 2017). However, land use patterns and changes significantly influence soil carbon sequestration capacity (Feller & Bernoux, 2008; Dindaroğlu et al., 2024).

Changes in land use have both direct and indirect effects on biotic and abiotic environmental factors within ecosystems (Dindaroğlu et al., 2024). As a result, land use changes are considered key indicators of land degradation and global climate change (Babur et al., 2021a; Dindaroğlu et al., 2021b). In recent years, improper land use and land use changes have become the primary drivers of both global and local environmental issues (Osman et al. 2025). Consequently, research on identifying, monitoring, and predicting future land use changes is growing rapidly. The conversion of forested areas into agricultural land and pastures negatively impacts land sustainability and ecosystem stability (Long & Liu, 2016). In contrast, afforestation of bare areas enhances soil and ecosystem protection (Babur et al., 2016). According to Bruce et al. (1999), deforestation and soil erosion lead to the release of approximately 1.7 and 0.1 PgCyr⁻¹ of carbon emissions into the atmosphere, respectively. However, increasing forest cover, rehabilitating degraded forests, implementing sustainable land management practices (Lal, 2001), and strengthening environmental protection measures (Wali et al., 1999; Kara et al., 2016; Babur et al., 2021a) can significantly mitigate global CO₂ emissions (Post & Kwon, 2000).

Lebanon cedar (*Cedrus libani* A. Rich.) forests are most densely distributed in the Taurus Mountains along Türkiye's Mediterranean coast (Boydak, 1996; 2003). Historically, extensive Lebanon cedar forests existed in Syria and Lebanon, but due to logging, burning, and goat-grazing over the past 5000 years, only small populations remain in these regions (Aytug, 1970). In the Taurus Mountains, Lebanon cedar generally thrives between 800 and 2100 meters in elevation, though smaller populations or individual trees can be found at lower (500–600 m) and higher (up to 2400 m) elevations. Additionally, scattered populations exist in other parts of Anatolia, such as Sultandağı-Afyon and the Black Sea region (Çatalan-Erbaa and Akıncıköy-Niksar) (Saatcioglu, 1976; Boydak, 1996; Atalay, 1987).

Currently, Türkiye hosts the world's largest natural Lebanon cedar forest, covering an area of 417,188 hectares (Çalışkan, 1998; URL-1, 2009). This species' resilience to extreme stress conditions and social pressures in the Mediterranean region, along with its high success rate in afforestation and reforestation, has made it a top choice for forest restoration efforts. Despite centuries of human activity in Anatolia, the rugged and inaccessible topography of the Taurus Mountains has played a crucial role in preventing the extinction of the Lebanon cedar (Boydak, 2003).

Inventory, estimation, projection, and management of soil and soil-related ecosystem resources in our country and the world require accurate determination of SOC stocks within the scope of management practices. In addition to the fact that a large part of the carbon pool in terrestrial ecosystems is soil (Jobbágy & Jackson, 2000), SOC is the main driver of critical ecosystem processes and services such as nutrient cycling, water retention, and biological carbon sequestration (Lal, 2004; Babur et al., 2021b). Although studies on C production and storage in aboveground ecosystem pools have intensified (Vance, 2000; Birdsey et al., 2006; Ludwig et al., 2011), the effects of management practices (such as afforestation, rehabilitation, harvesting) on the increase of belowground C stocks at temporal and spatial scales are much less understood (An et al., 2009). Although various articles have been published on C sequestration in soils in humid and tropical regions, studies on C sequestration are needed in Türkiye since it is covered with semiarid regions with widespread soil degradation, is under the influence of extreme ecological conditions, and is exposed to the negative effects of climate change. Therefore, this study was carried out to reveal how cedar afforestation carried out at different times in karst areas, which are sensitive as ecosystems, for soil protection against erosion, changes the C and N stocks stored in the soils over time. The data to be obtained will reveal the amount of C stored in bare karst areas after afforestation and the function of afforestation, which plays an effective role in reducing greenhouse gas emissions that play a role in global climate change.

MATERIAL AND METHODS

Study Site

The research area consists of afforestation sites within the borders of Toros and Sorgun villages in the Erdemli district of Mersin province, located in the Mediterranean Region of Türkiye (Figure 1). Erdemli's geographical coordinates are 36°34' N latitude and 34°18' E longitude. The district has a warm and temperate climate, with more rainfall in winter than in summer. According to the Köppen–Geiger classification, Erdemli falls under the Cold Semiarid (Csa) climate category, characterized by mild winters and very hot, dry summers. The district's annual average temperature is 16.1 °C, with August being the hottest month (25.5 °C) and January the coldest (6.9 °C). The annual total rainfall is 650 mm, with December receiving the highest precipitation (140mm) and July the lowest (15mm).



Figure 1. Location of research sites

Given the high-altitude nature of the study area, afforestation efforts have focused on Taurus cedar (*Cedrus libani* L.), which thrives at these elevations. The predominant soil type is Terra rossa (Previtali et al., 2017). Geologically, the study area is part of a basin where Tertiary sedimentary rocks from the Oligocene-Pliocene period rest unconformably on the rugged topography of Paleozoic-Mesozoic basement rocks forming the Taurus orogenic belt (MTA, 2020). These durable limestone surfaces have formed steep slopes, featuring numerous karst dissolution cavities, slides, and rock formations, particularly in the uppermost sections of the terrain (Bulut, 1998). Some physiographic, topographic, biotic and edaphic features of the research area are shown in Table 1.

Table 1. Study site characteristics				
Environmental Factors	Afforestation Ages			
	Control	10 years	25 years	
Longitude	36° 54' 13''	36° 54' 22''	36° 49' 55''	
Latitude	34° 06' 52''	34° 06' 37''	34° 97' 49''	
Altitude mean (m)	1670	1675	1650	
Slope degree mean (%)	30	40	40	
Afforestation date	0	2015	2000	
Vegetation type	-	Taurus cedar	Taurus cedar	
Soil texture	Sandy clay loam	Sandy clay loam	Sandy loam	
pН	7.01	8.09	6.80	
pH class	Neutral	Slightly Alkaline	Neutral	

Soil Sampling and Laboratory Analysis

Soil samples were collected in October 2024 from afforested in different years, as well as from non-afforested bare areas (control) within the borders of the Erdemli Forest Management Directorate. A total of 45 sampling points were randomly selected: 15 from each different cedar afforestation area established in 2000 (C25) and 2015 (C10), and 15 from

control sites (Co). From these points, 45 disturbed and 45 undisturbed soil samples were collected from topsoil layer (0-10 cm). To determine bulk density, undisturbed soil samples were taken using a sample ring (steel cylinder=VS= 385cm³), while disturbed soil samples were collected separately for chemical analyses. Soil sampling in forested areas followed the International Cooperative Programme (ICP) Guidelines (UNECE, 2003) and the Area-Frame Random Soil Sampling (AFRSS) methodology (EC, 2009; IPCC, 2003; Stolbovoy et al., 2007)

The bulk density of the soil samples with intact natural structure (undisturbed soil) brought to the laboratory was calculated after they were dried in an oven for 24 h at 105 °C until they reached a constant mass weight and weighed on a precision scale (Sariyildiz et al., 2024; Dindaroğlu et al., 2024).

Soil organic carbon (SOC) and total nitrogen (TN) contents were analyzed using the Walkley-Black chromic acid wet oxidation method and the Kjeldahl digestion method, respectively (Rowell, 1994).

SOC and TN stocks were calculated according to the Formula (1) given below (Lee et al., 2009).

SOC or
$$TN_{stocks} = \% SOC$$
 or $\% TN * Mi$ (ton ha⁻¹) (1)

Mi given in this formula represents the mass of dry soil at the i'th soil depth and its amount is calculated according to the below Formula (2).

Mi: BDi x Ti x 10⁴

BDi represents the soil bulk density at i soil depth (ton m^{-3}), Ti represents the soil sampling thickness at i soil depth (m), and 10^4 represents the unit change factor (m^2 ha⁻¹).

Statistical Analysis

Descriptive statistical analyses were made regarding the BD, SOC, TN, C_{stock} , and TN_{stock} properties of soils taken from cedar-afforested and control lands at different times. The Kolmogorov-Smirnov analysis method was used to test whether the values obtained as a result of the analyses showed a normal distribution based on the afforestation year. Duncan's test from one-way variance analysis was used to compare independent group differences. The statistical significance level was accepted as P<0.05.

RESULTS

Descriptive statistics of some soil properties that play an active role in the decomposition processes in Control (Co), 10-year-old cedar field (C10), and 25-year-old cedar field (C25) land use areas are presented in Table 2. BD values ranged from 0.49 to 1.54 g/cm³ in C25, 0.68 to 1.64 g/cm³ in C10, and 0.83 to 1.94 g/cm³ in Co. SOC content varied between 2.08% and 6.46% in C25, 1.35% and 5.80% in C10, and 0.32% and 2.40% in Co. TN levels ranged from 0.015% to 0.153% in C25, 0.037% to 0.108% in C10, and 0.010% to 0.059% in Co.

(2)

Soil carbon stock (C_{Stock}) values fluctuated between 15.01 and 52.47 tC ha⁻¹ in C25, 13.69 and 50.72 tC ha⁻¹ in C10, and 3.13 and 39.54 tC ha⁻¹ in Co. Similarly, soil nitrogen stock (N_{Stock}) varied between 0.190 and 1.15 tC ha⁻¹ in C25, 0.380 and 1.55 tC ha⁻¹ in C10, and 0.130 and 1.14 tC ha⁻¹ in Co.

		N	Maan	Std Deviation	Std Eman	Minimum	Movimum
		IN	Ivican	Stu. Deviation	Std. Elloi	WIIIIIIIII	Waxiiiiuiii
	C25	15	1,07	,27	,069	,49	1,54
BD	C10	15	1,13	,24	,061	,68	1,46
	Со	15	1,35	,3	,081	,83	1,94
	C25	15	3,35	1,23	,316	2,08	6,46
SOC	C10	15	2,86	1,19	,309	1,35	5,80
	Со	15	1,55	,75	,194	,32	2,40
	C25	15	,051	,035	,009	,015	,153
TN	C10	15	,070	,021	,006	,037	,108
	Со	15	,036	,017	,004	,010	,059
	C25	15	34,97	11,44	2,954	15,01	52,47
C _{Stock}	C10	15	31,19	10,91	2,816	13,69	50,72
	Со	15	21,13	11,50	2,970	3,13	39,54
	C25	15	,51	,28	,073	,190	1,15
N _{Stock}	C10	15	,78	,27	,070	,380	1,55
	Co	15	,49	,28	,072	,130	1,14

 Table 2. Descriptive Statistics of Topsoil (0–10 cm)

Abbreviations: BD=Bulk Density (g/cm³); SOC= Soil organic carbon; TN= total nitrogen; C_{Stock} =soil organic carbon stock (tC ha⁻¹); N_{Stock}=soil organic nitrogen stock (tN ha⁻¹) C25= 25 ages cedar forest; C10= 10 ages cedar forest; Co=Control

ANOVA tests conducted on measured soil samples and the average values of carbon (C) and nitrogen (N) stocks indicated that afforestation significantly influenced these soil properties over time ($P \le 0.05$; Tables 3 and 4). Among these properties, SOC exhibited the highest F-value.

Table 3. *F and P* statistic values of the one-way ANOVA for temporal effect of afforestation on BD, SOC, TN, and C and N stocks

	BD	SOC	TN	Cstock	Nstock
P-values	*	***	**	**	**
F-values	4.25	11.13	6.64	6.03	5.14

Abbreviations: BD=Bulk Density (g/cm³); SOC= Soil organic carbon; TN= total nitrogen; C_{Stock}=soil organic carbon stock (tC ha⁻¹); N_{Stock}=soil organic nitrogen stock (tN ha⁻¹), * Significant at $P \le 0.05$; ** Significant at $P \le 0.01$; *** Significant at $P \le 0.001$.

Table 4. According to the one-way ANOVA, the changes in soil properties with afforestation of bare space over different years.

		Afforestation Ages	
Properties	Co	C10	C25
BD (g/cm^3)	1.35±0.31 ^b	1.13±0.24 ^a	1.07±0.27 ª
SOC (%)	1.55±0.75 °a	2.86±1.19 ^b	3.35±1.23 ^b
TN(%)	.036±0.02 ª	.070±0.02 ^b	$.051{\pm}0.04^{\text{ ab}}$

The mean BD values of the soils were found to be different ($P \le 0.05$; Table 3). The highest BD was observed in control soils, while the lowest was recorded in C25 soils.

The organic C fraction in the soil is significantly related to the age of afforested sites (Tables 3 and 4). The SOC values in soils from cedar afforestation sites were higher than those from bare land, with a statistically significant difference between afforested and bare land areas. Especially, C25 soils had the highest C levels, whereas control soils had the lowest, showing an inverse relationship with BD. Total nitrogen content was highest in the C10 and lowest in Co soils.

The average organic carbon sequestration of soils was calculated and the highest value was found in C25 (34.97 tC ha⁻¹), while the lowest was recorded in Co soils (21.13 tC ha⁻¹), mirroring the pattern observed in soil C content (Figure 2a). Similarly, N stocks were highest in C10 (0.78 tN ha⁻¹) and lowest in Co soils (0.49 tN ha⁻¹), aligning with TN content trends (Figure 2b). In terms of C_{Stocks}, C25 and C10 soils formed distinct groups from control soils. However, for N stocks, C10 soils exhibited significant differences compared to both control and C25 soils.



Figure 2. Changes in soil organic carbon stocks (a), total nitrogen stocks (b), with afforestation in different years. Different letters above the bars indicate significant differences at p < .05 among the land use types.

The Pearson correlation analysis illustrated that there was a negative significant relationship between BD, SOC, and TN (p<0.01 and p<0.05). In addition, there was a significant positive relationship between SOC and TN, C_{Stock} and N_{Stock} (p<0.01). The highest relationship was found between TN and N_{Stock} (p<0.01; r=0.845**) (Figure 3).



Figure 3. The heatmap of the Pearson correlation coefficients (p<0.05).

DISCUSSION

Afforestation practices significantly influenced the BD, OC, and TN values in the research area. The highest BD of the research areas was found in the control areas. A significant increase in BD values was found in the 0–10 cm soil layer, corresponding to the age of afforestation areas. The highest BD values were recorded in bare land soils (Tables 2 and 3). This increase in BD in bare lands can be attributed to lower soil organic matter content, degradation of soil structure, and a reduction in macropores due to soil deterioration. Similarly, Yuksek & Yuksek (2011) reported the highest BD (1.32 g/cm³) in bare land and the lowest BD (1.18 g/cm³) in the upper layers of afforested soils. Our study site was on Karst ecosystems. Previous studies on karst ecosystems by Vermez et al. (2018) indicated that soils were moderately basic (pH 8.1) and contained sufficient organic matter (4.33%).

Generally, afforested lands exhibited higher OC and TN values than bare lands (Tables 2 and 3). This increase is attributed to the accumulation of dead needles, branches, and cones, which enhance soil carbon and nitrogen content by supplying organic matter. The primary sources of C and N in forest soils are fallen litter and fine roots (Babur et al., 2021a). The quantity and quality of litter contribute to plant primary productivity by enhancing fine root density and turnover, thereby increasing SOC and TN availability (Laik et al., 2009; Pang et al., 2016).

Similar findings have been reported in previous studies. Lima et al. (2006) found that afforestation of degraded pastures led to increased carbon accumulation in soils over 30 years. Afforestation has been shown to improve various soil properties, including physical, hydrophysical, and chemical characteristics (Yuksek & Yuksek, 2011; Kara et al., 2016). For example, organic matter content in soils with Black Locust plantations ranged between 2.09%

and 3.57%. However, Polglase et al. (2000) noted that afforestation only had a minor effect on soil carbon accumulation across different environments.

Conversely, some researchers observed a temporary decline in total soil organic carbon after converting grasslands to forests, with reductions appearing a few years after the land-use change (Thuille & Schulze, 2006; Alberti et al., 2008). This trend is attributed to the higher fine root biomass in natural grasslands compared to plantation sites (Guo et al., 2007), as fine roots are a primary source of SOC (Carter & Gregorich, 2010; Babur et al., 2021b). In contrast, bare lands experience substantial soil and organic carbon loss due to erosion and topsoil displacement (Babur et al., 2016). To enhance OC stock in bare lands, urgent measures such as grazing management, rehabilitation, or afforestation are necessary. In bare lands, the absence of protective vegetation results in lower carbon inputs to the topsoil. Additionally, erosion-induced organic matter loss further contributes to reduced SOC levels.

In our study, the correlation matrix indicated a negative relationship between SOC and BD, with BD increasing as SOC decreased (Table 4). As SOC increased, BD decreased. This inverse relationship occurs because SOC enhances soil porosity, thereby reducing bulk density (Korkanç, 2014). Korkanç (2014) also stated that 1.58 g/cm3 BD, 1.49% organic carbon, and accordingly 23.54 tC ha-1 carbon were stored in the topsoil layer (0-10cm) of 14-year cedar afforestation areas. Additionally, Dindaroğlu et al. (2024) reported that forested areas exhibited the highest soil carbon stocks, reaching 101.56 tC ha⁻¹ in the top 30 cm of soil. It has also been reported that maintenance work in afforestation areas significantly increases the SOC stocks of soils by 7.2% (Gong et al., 2021).

CONCLUSION

This study concluded that land use and land cover changes in degraded lands may affect some important soil properties. In particular, afforestation activities carried out in degraded lands have shown that they positively affect the determined soil properties (SOC, TN, and bulk density). In addition, it is predicted that soil quality may further increase with age in terms of these properties. Afforestation activities to be planned using cedar saplings for afforestation of high-altitude karst areas in the Northern Mediterranean will have a positive effect on soil carbon sequestration and will help reduce the effects of global warming. As a result of afforestation activities carried out in degraded lands of this semiarid Mediterranean coastline, which is sensitive to the effects of climate change, soil organic carbon stocks have increased significantly; This situation may be useful in determining which tree species to plant in the future afforestation activities to combat the effects of global warming.

AUTHOR CONTRIBUTIONS

All authors contributed equally to the article. There is no conflict of interest.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

ETHICS COMMITTEE APPROVAL

This study does not require any ethics committee approval.

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