# **Supplementary materials**

Role of photosynthesis and stomatal conductance on the long-term rising of intrinsic water use efficiency in three old-growth forests in Bosnia-Herzegovina and Montenegro

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Figure S1 – Location of the three experimental sites.

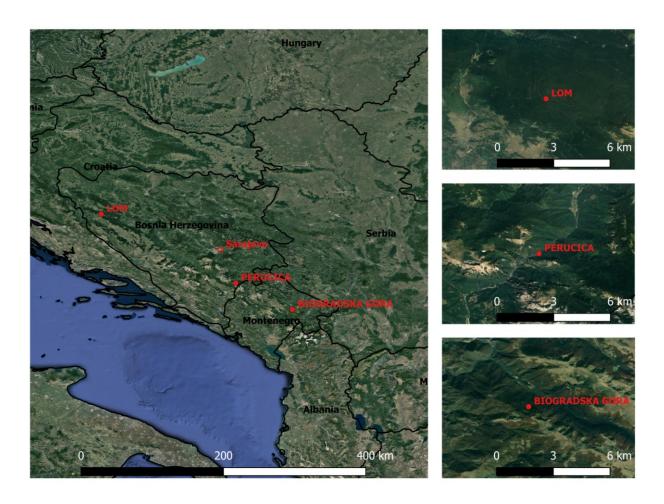


Figure S2 – Distribution of the standing trees by diameter and height class within the 1-ha permanent plot in Perucica (two panels on the top) and relative distribution of the sampled fir trees by diameter and height class in the same plot (two panels on the bottom).

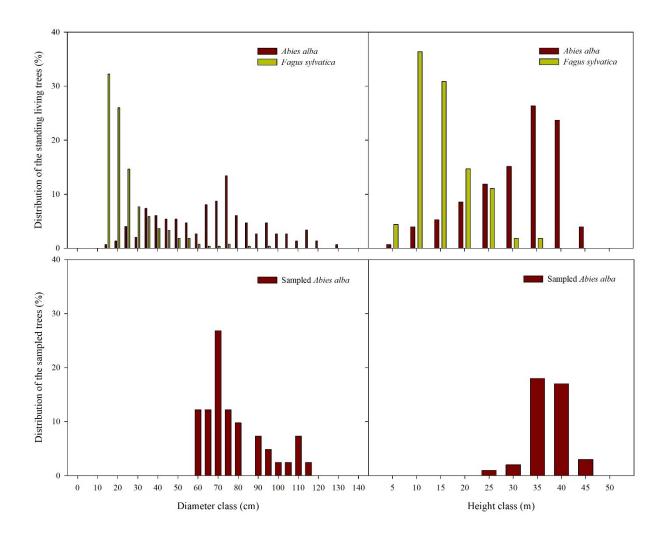


Figure S3  $-\delta^{18}O$  of the precipitation estimated following Barbour et al. (2001) (Equation 8) at the weather station in Sarajevo (top) and in Zagreb (bottom).

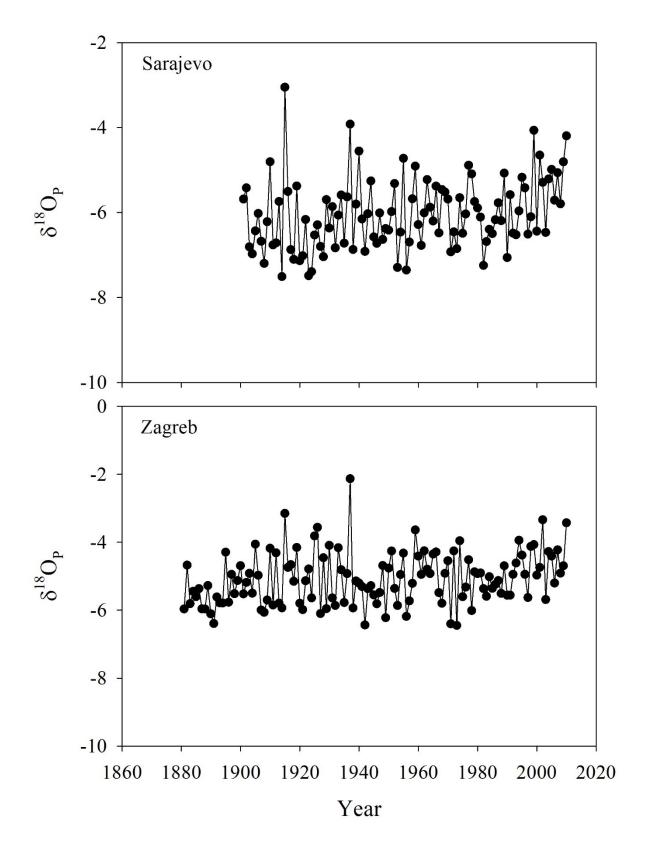


Figure S4 – Ratio between intercellular CO<sub>2</sub> concentration ( $C_i$ ) and atmospheric CO<sub>2</sub> concentration ( $C_a$ ) by selected decades at the three old-growth forests (PER = Perucica; LOM = Lom; BIO = Biogradska Gora). Mean  $\pm$  standard error. Different letters indicate significant differences among periods (p<0.05).

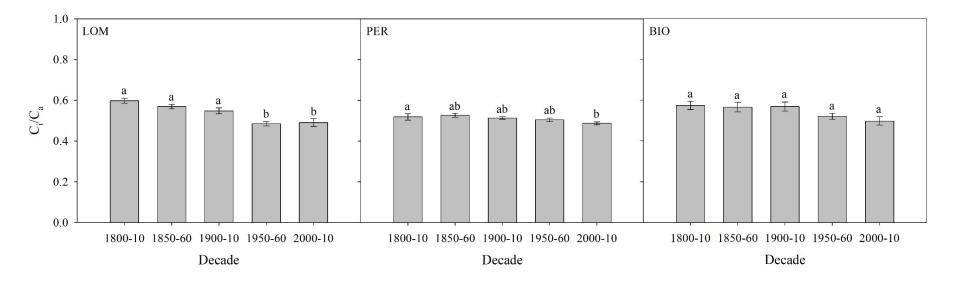


Table S1 - ANOVA results for intercellular  $CO_2$  concentration  $C_i$  by selected decades (see Figure 1 in the main text) at the three old-growth forests (PER = Perucica; LOM = Lom; BIO = Biogradska Gora).

Source of Variation	DF	SS	MS	F	P
Between Groups	4	3680.201	920.050	7.348	< 0.001
Residual	25	3130.415	125.217		
Total	29	6810.616			

### <u>PER</u>

<b>Source of Variation</b>	DF	SS	MS	$\mathbf{F}$	P
Between Groups	4	5590.168	1397.542	35.549	< 0.001
Residual	22	864.900	39.314		
Total	26	6455.069			

## <u>BIO</u>

Source of Variation	DF	SS	MS	F	P
Between Groups	4	3298.950	824.738	4.071	0.014
Residual	20	4051.613	202.581		
Total	24	7350.563			

Table S2 - ANOVA results for basal area increment (BAI) by selected decades (see Figure 2 in the main text) at the three old-growth forests (PER = Perucica; LOM = Lom; BIO = Biogradska Gora).

Source of Variation Between Groups Residual Total	<b>DF</b> 4 25 29	SS 1163.229 644.382 1807.611	<b>MS</b> 290.807 25.775	<b>F</b> 11.282	<b>P</b> <0.001
<u>PER</u>					
Source of Variation Between Groups Residual Total	<b>DF</b> 4 25 29	<b>SS</b> 836.463 992.303 1828.765	MS 209.116 39.692	<b>F</b> 5.268	<b>P</b> 0.003
BIO Source of Variation Between Groups Residual Total	<b>DF</b> 4 20 24	SS 3697.236 2710.165 6407.402	MS 924.309 135.508	<b>F</b> 6.821	<b>P</b> 0.001

Table S3 – ANOVA results for intrinsic water use efficiency (iWUE) by selected decades (see Figure 2 in the main text) at the three old-growth forests (PER = Perucica; LOM = Lom; BIO = Biogradska Gora).

LOM Source of Variation Between Groups Residual Total	<b>DF</b> 4 25 29	SS 19729.437 2519.164 22248.600	MS 4932.359 100.767	<b>F</b> 48.948	<b>P</b> <0.001
PER Source of Variation Between Groups Residual Total	<b>DF</b> 4 22 26	SS 9693.210 695.356 10388.566	MS 2423.303 31.607	<b>F</b> 76.670	<b>P</b> <0.001

<u>BIO</u> Source of Variation	DF	SS	MS	F	P
Between Groups	4	13063.888	3265.972	19.987	< 0.001
Residual	20	3268.105	163.405		
Total	24	16331.993			

Table S4 – ANOVA results for leaf water evaporative enrichment ( $\Delta^{18}O_L$ ) by selected decades (see Figure 2 in the main text) at the three old-growth forests (PER = Perucica; LOM = Lom; BIO = Biogradska Gora).

## <u>LOM</u>

<b>Source of Variation</b>	DF	SS	MS	F	P
Between Groups	2	3.855	1.927	1.080	0.365
Residual	15	26.773	1.785		
Total	17	30.628			

## <u>PER</u>

<b>Source of Variation</b>	DF	SS	MS	$\mathbf{F}$	P
Between Groups	2	0.510	0.255	0.0725	0.930
Residual	15	52.711	3.514		
Total	17	53.221			

## <u>BIO</u>

<b>Source of Variation</b>	DF	SS	MS	F	P
Between Groups	2	3.720	1.860	0.756	0.491
Residual	12	29.535	2.461		
Total	14	33.255			

Table S5 – ANOVA results for the ratio between intercellular  $CO_2$  concentration ( $C_i$ ) and atmospheric  $CO_2$  concentration ( $C_a$ ) by selected decades (see Figure S4) at the three old-growth forests (PER = Perucica; LOM = Lom; BIO = Biogradska Gora).

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<b>Source of Variation</b>	DF	SS	MS	F	P
Between Groups	4	0.0582	0.0145	12.665	< 0.001
Residual	25	0.0287	0.00115		
Total	29	0.0869			

## <u>PER</u>

<b>Source of Variation</b>	DF	SS	MS	$\mathbf{F}$	P
Between Groups	4	0.00509	0.00127	3.079	0.037
Residual	22	0.00910	0.000414		
Total	26	0.0142			

### <u>BIO</u>

<b>Source of Variation</b>	DF	SS	MS	$\mathbf{F}$	P
Between Groups	4	0.0231	0.00578	2.748	0.057
Residual	20	0.0421	0.00210		
Total	24	0.0652			

Table S6 – Regression results of figure 4 (PER = Perucica; LOM = Lom; BIO = Biogradska Gora).

#### <u>LOM</u>

R: 0.8217 R<sup>2</sup>: 0.6752 Adj R<sup>2</sup>: 0.6636

Standard Error of Estimate: 16.0645

	Coefficient	Std. Error	t	P
y0	86.4602	5.2475	16.4765	< 0.0001
a	2.8828	0.3778	7.6297	< 0.0001

## Analysis of Variance:

	DF	SS	MS
Regression	2	444571.4845	222285.7422
Residual	28	7225.9309	258.0690
Total	30	451797.4154	15059.9138

#### Corrected for the mean of the observations:

	DF	SS	MS	$\mathbf{F}$	P
Regression	1	15022.6695	15022.6695	58.2118	< 0.0001
Residual	28	7225.9309	258.0690		
Total	29	22248.6005	767.1931		

#### <u>PER</u>

R: 0.6462 R<sup>2</sup>: 0.4175 Adj R<sup>2</sup>: 0.3942

Standard Error of Estimate: 15.5579

	Coefficio	ent Std.	Error t	P	
y0	112.0536	4.9184	22.7825	< 0.0001	
a	1.6031	0.3787	4.2331	0.0003	
Analysis of Va	riance:				
·	DF	SS	MS		
Regression	2	450664.1382	225332.0691		
Residual	25	6051.2229	242.0489		
Total	27	456715.3611	16915.3837		
Corrected for the	he mean of the	e observations:			
	DF	SS	MS	${f F}$	P
Regression	1	4337.3431	4337.3431	17.9193	0.0003

Residual	25	6051.2229	242.0489
Total	26	10388.5660	399.5602

# <u>BIO</u>

R: 0.6722 R<sup>2</sup>: 0.4519 Adj R<sup>2</sup>: 0.4280

Standard Error of Estimate: 19.7286

	Coefficient	Std. Error	t	P
y0	98.3622	5.8708	16.7544	< 0.0001
a	0.8524	0.1958	4.3544	0.0002
Analysis of	f Variance:			

	DF	SS	MS
Regression	2	351313.2271	175656.6136
Residual	23	8952.0094	389.2178
Total	25	360265.2365	14410.6095

### Corrected for the mean of the observations:

	DF	SS	MS	$\mathbf{F}$	P
Regression	1	7379.9833	7379.9833	18.9611	0.0002
Residual	23	8952.0094	389.2178		
Total	24	16331.9927	680.4997		