Research Article

Climate Change and Plant Anatomy: Assessment of Adaptability Potential of Walnut *(Juglans regia)* Grown in Swat, Pakistan

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Abstract | Trees specie s as living entity response to climate and record variations in climate; thus are commonly used to study climate change. The aim of this study was to explore the dendrochronological and dendroclimatological potential of broad-leaved species, specifically Walnut (Juglans regia) in dry tem perate fore st zone, Swat, Pakistan. Juglansregia was selected to assess the resilience and adaptability potential of the species in the scenario of climate change, because no broad-leaved species have been studied so far in dry temperate forest in Swat region to establish climate-growth relationship. To that objectives, chronologies and time series were developed of annual rings and vessel areas of the species. The results revealed the high degree of annual variations in the species due to climatic fluctuations. Dendrochronological potential of the species is found moderate thus the chronology may be used as substitute to study climatic variations. Tree ring width (TRW) and climate relationship was found relatively significant showing limitations of climate in the radial growth. Precipitation found as a strong growth limiting factor in comparison to temperature. Beyond the year 2000 C.E, the rainfall ratio was found to be on the decline, whereas the tem perature was on the rise; this phenomenon triggered fast radial growth. Moreover, increase in the diameter of the vessels was observed in the species more probably as a result of rise in mean annual temperature and decline in the precipitation in the area.

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Introduction

The earth's life-sustaining systems are deteriorating due to significant climate changes (Hardy, 2003), causing unprecedented weather patterns and environmental occurrences never seen before (Lipczynska-Kochany, 2018; Change, 2018; Hegerl *et al*., 2007). Human activities, leading to greenhouse gas emissions, are disrupting the natural climate change process and resulting in rising air temperatures, elevated ocean levels, and glacier movement and melting (Nicholls *et al*., 1999;

Marchant and Hooghiemstra, 2004; McCarroll and Loader, 2004). Globally, the poleward migration of various genera and ecosystems is occurring due to widespread global warming, a phenomenon outlined by Cramer *et al.* (2014). This warming is responsible for the emergence of exceptional weather events and repetitive disruptions to seasonal patterns as early flowering in plants (Settele *et al.*, 2015). It is expected that earth's temperature may rise by 1.5-2.0°C by the end of 21st century, and the climatic research studies reveals potential consequences such as a 20-30% extinction rate among plant species (Edenhofer, 2015).

Pakistan's vulnerability to environmental changes is evident; most of the Asian countries including Pakistan facing urbanization, industrial progress and resource exhaustion these factors are limiting ecological development and sustainability (Chan *et al.*, 2018; Shaffril *et al.*, 2018). Pakistan has recently faced unparalleled intense weather events resulting in crop diseases and evident seasonal changes (Abid *et al.*, 2016; GoP, 2014, 2017; Hussain *et al.*, 2018); moreover, agriculture sector and limited forest cover is in decline in Pakistan generally and in hot moist areas specifically (Abas *et al.*, 2017; Olsen, 2009; FAO, 2020). The common masses' awareness of adaptable climate approaches is necessary to avoid bitter consequences (Perkins *et al.*, 2018).

This research was initiated to investigate precipitation, temperature data and trends in the Swat; and the impact of shifts in temperature and precipitation in wood anatomy of Walnut and its adaptability potential. Moreover, it may help in developing a model to predict future climatic impact and changes in the anatomy, adaptability, drought resistance and survival potential of Walnut grown in Swat over the next few decades. This study may help in developing policies in local socioeconomic context of Swat region in terms of plantation of the walnut trees.

In Pakistan the impact of climate change in the anatomy of hardwood species has never been studied; thus, there is big gap of knowledge and need to be filled. Therefore, it is need of the day to evaluate the sensitivity and response of trees species toward climate to determine their adaptability potential against climatic happenings. This is necessary to preserve biodiversity for future generation.

The objectives of this research are: (1) to investigate the response of the anatomical feature toward climatic; (2) to assess the sensitivity and adaptability potential of the species in Swat region in this changing phenomena of the environment.

Materials and Methods

Description of study site

Swat is located in KP, Pakistan; the average elevation of Swat is 980 m (Mohiuddin and Niaz, 2007). Swat's lower reaches have vegetation characterized by dry bush and deciduous trees, while the upper areas mostly have thick pine forests (Barth, 2020). In Swat, besides other soft and hardwood species, *Juglans regia, Morus alba, Platanus orientalis, Salix tetrasperma and Melia azedarach* are common species. Swat falls in the zone of coniferous sub-alpine forests at the upper-most tree formation in Himalayas. *Abies pindrow, Pinus wallichiana* stand singly or in groups with broadleaved trees (Sheikh, 1993).

Description of species

Walnut (*Juglans regia*) is a large, slow grown, deciduous tree; found in northern areas including Swat. It is a moderately tolerant tree that grows on deep, rich, moist soils, but prefers moist, shady sites. Its precipitation zone is 750 to 1500 mm/yr with a temp: range of -10 to 35°C at elevations between 1000 and 3300 m (Sheikh, 1993). Wood is semi-ring-porous. Growth rings generally distinct and delimited by a narrow, sharp line of fibers. Vessels are generally large to medium-sized and small (Pearson and Brown, 1932).

Field and laboratory methods

For this study 10 walnut trees were selected from different location of Swat by using random sampling method and discs of full girth from the selected trees were collected. After drying, all discs were successively sandpapered with different grit sizes to gain the shine, finish and clarity. Discs of best ring and band parenchyma visibility were scanned at the HP LaserJet Pro MFP M26 nw at 1200 DPI and images were generated. The generated images were analyzed using tree measuring software CDendro and CooRecorder 9 to measure the annual rings (Maxwell and Larsson, 2021). Computer programs cofecha and ARSTAN were used for cross dating and standardization of the time series. Samples were cross dated and cofecha was used to verify cross dating (Stokes and Smiley, 1968; Holmes, 1983; Grissino-Mayer, 2001). After cross dating each ring was assigned a calendar year and final chronology of *Juglans regia* consists of 7 series. ARSTAN was used for standardization of the tree ring series (Cook, 1985). Hugershoff growth curve fitting was used for the standardization of the time series with the help of dependent smoothing spline (Melvin et al., 2007). Standardized series were combined for developing a signal free chronology by applying biweight robust mean to curtail the impact of outliers (Cook, 1985; Cook and Kairiukstis, 2013). Four chronologies i.e., Raw, Standard and ARSTAN and residual chronologies were developed by using ARSTAN. In this study residual chronology was used for establishing correlation with climatic factors. Schweingruber *et al.* (1988) recommends for developing a chronology minimum 10 trees must be sampled; thus 10 samples were cross dated to develop robust chronology.

Description of wood vessels

Wood vessels are the basic unit of the hardwood species; main function of the vessels is to conduct water and minerals from soil to the upper part of the plants. The anatomy and geometry of the vessels may change with availability of the water. Thus, the diameter of the vessels was selected to assess the response of the species toward climate change. For studying vessel diameter for the assessment of the climatic impact on diameter of vessels; a radial strip was cut off from the main discs of the samples. These strips were converted into blocks of one (1) cubic inch; blocks were kept for three days in boiling water for softening. After softening of blocks the microsection of 30µ were prepared on sledge microtome. These sections were properly prepared, stained under a specific process and mounted on slides for optical analysis and collection of data.

Descriptive statistics and analysis

Descriptive statistics as interseries correlation, mean sensitivity, correlation coefficients, Gleichläufigkeit (GLK) and student's t-test were calculated. Interseries correlation was measured to gauge the strength of cross dating for as site, mean sensitivity values to check the sensitivity that requires a value above 0.30 which represents the sensitive measurement with high standard deviation and low autocorrelation (Grissino-Mayer, 2001). Correlation coefficients, Gleichläufigkeit and t-values were calculated to validate the cross dating. Gleichläufigkeit (GLK) and Student's t-test were used as the key parameters applied to scale a successful cross dating. Recommended threshold for student's t-test is t-value ≥ 1.96 and GLK values ≥ 70 (Von Platen and Eker, 2008) were applied.

Results and Discussion

Climatic variations play a crucial role in the development and survival of the biodiversity of flora. Trees always vulnerable to climate and sensitive to variations in the climate as the climate is major element that may limit the growth of trees. Thus, climate and climatic factors assumed as major controlling players when studying the limiting tree growth factors (Figure 1, Table 1).

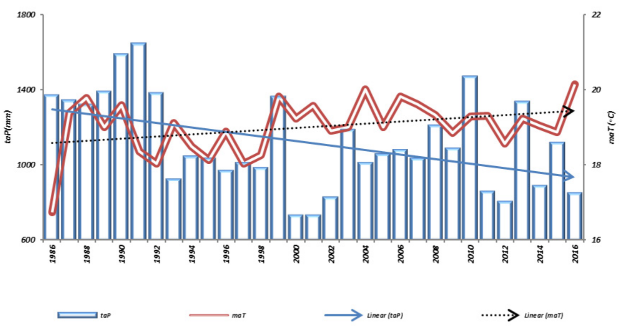

Figure 1: *Climatic indices of swat.*

Table 1: *Statistics of climatic parameters of Swat.*

Zone	Climatic parameters	Descriptive statistics			Decadal variations				Post 2000 variations	
		MV	Max	Min	Decades	MV	Increase	Decrease	Increase	Decrease
Swat	Precipitations (mm)	1113.92	1639.9	728.2	1986-1996	1267	-	-	-	-171.64
					1997-2006	994.2	-	-275.11		
					2007-2016	1063	68.39	-		
	MAT (°C)	19.01	20.15	16.73	1986-1996	18.68	-	-	0.56	-
					1997-2006	19.17	0.49	-		
					2007-2016	19.22	0.05	-		

Source: *Pakistan Meteorological Department.*

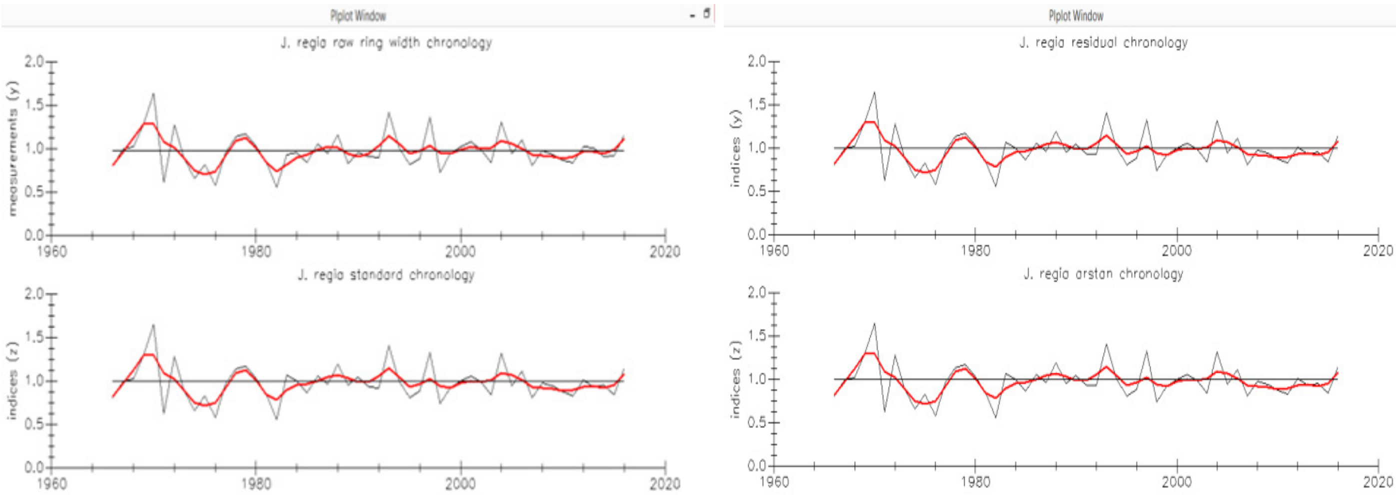

Figure 2: *ARSTAN generated chronologies of Juglans regia grown Swat.*

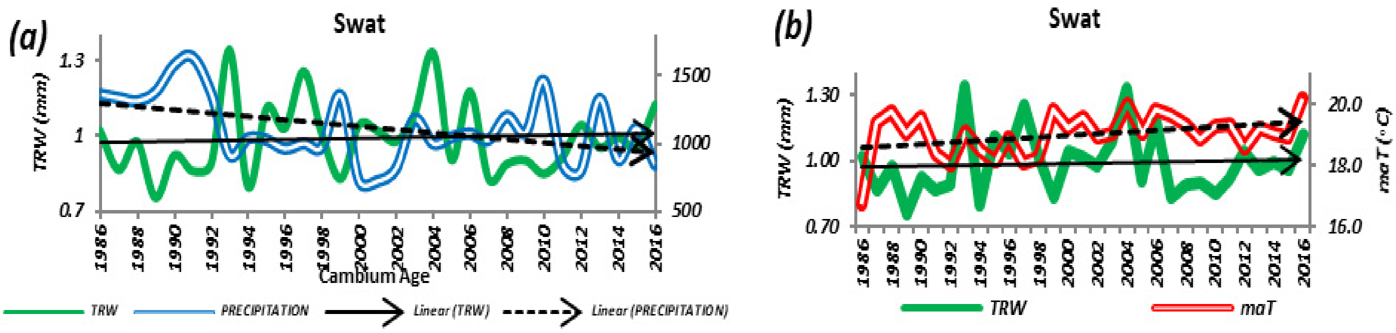

Figure 3: *(a) TRW and TAP (b) TRW and MAT.*

Descriptive statistics

Analysis of the studied species revealed high mean value in tree ring width. CV% shows that species expressed moderate heterogeneity of the ecological and climatic conditions. Mean sensitivity result displays that the species shows the higher ring sensitivity (Table 2). This means that Swat experiences the high degree of annual climatic variations.

Dendrochronological prospective

Results revealed high correlation coefficients, t values and *Gleichläufigkeit* score, which shows that there is highest degree of match between the sequences observed in the species grown in Swat. That means the cross-dating is good enough in the species. Furthermore, moderate SI and MS values show that cross-dating is moderately successful and the species can be used for chronology in Swat. Low value of autocorrelation expresses that the TRW sufficiently affected by external factors rather than endogenous. The species is suitable to study climate in Swat (Figure 2).

TRW-climate relationship

Results revealed the strong and positive relationship between the tree ring indices and climatic variables; in tree ring indices up to 5.5% variance has been explained by the climate in general. Results revealed that climate have impact on growth the species grown Swat.

Moreover, results revealed negative but significant precipitation-TRW relationship; values of R^2 shows variance up to 4% tree ring width has been explained by rainfall in Swat region. Beta values reveals an increase in rainfall indices by 01 mm could cause an incitation in growth of the species up to 0.0004 mm radial growth (Table 2); however, TRW-rainfall relationship was found non-significant.

Moreover, TRW-MAT relationship was found weak and negative in Swat region. Regression analysis shows 0.01% variance in the tree rings width which have been explained by the MAT. Beta values revealed that rise in MAT by the value of 01 Celsius may cause a decrease in tree ring width up to -0.004 mm. It was found that rise in mean annul temperature may restrict radial growth of species (Figure 3).

Decadal variations in TRW

Decadal calculations indicated fast growth rate in species in 2nd decade (1997-2006) along with decrease in precipitation and rise in temperature (Table 1). Relatively low precipitation ratio along with high temperature triggered fast radial growth in the species.

Variations in TRW after year 2000

Fast growth in the species after year 2000 was observed along with decline in rainfall however rise in temperature (Table 1). Results revealed that decrease in rainfall and rise in MAT caused high growth rate after year 2000.

Anatomy and dendrochronological perspective

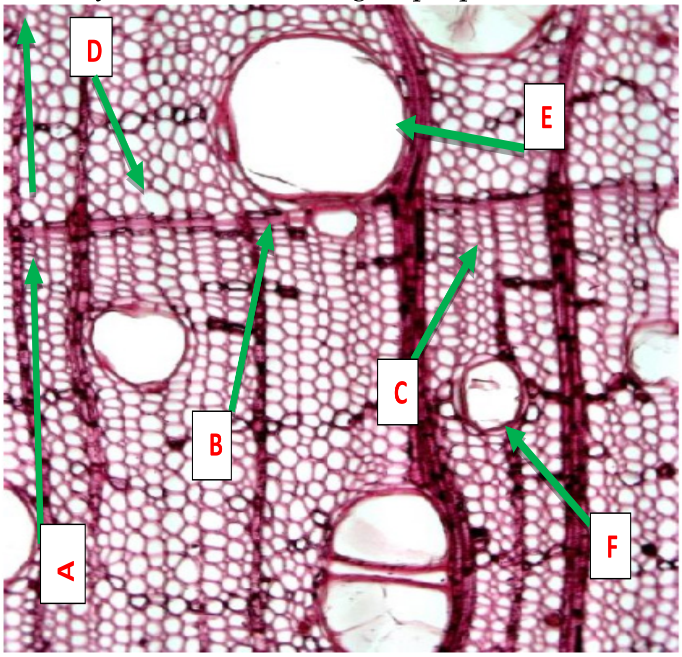

Figure 4: *(A) Early to late wood growth (B) Annual ring border line (C) Small diameter of representing late wood (LW) (D) Large diameter of fiber representing early wood (EW) (E) EW vessel (F) LW vessels.*

Walnut wood observed as semi ring porous, early wood vessel and fiber geometry and dimensions are comparatively different from the late wood. Early wood vessels and fiber diameter are large enough in comparison to late wood (Figure 4). Thus, counting and measurements of annual rings was relatively easy and this is necessary assessing dendroecological and dendroclimatological potential and assessing the adaptability of the species.

Mean frequency of the vessels were found up to 13 mm² (Table 2). Weak but positive association was found between the total annual precipitation (TAP) and vessel diameter of the species. TAP-vessel diameter relationship was found non-significant

Table 2: *Comparison of impacts of climate change on TRW and vessel of Juglans regia.*

Zone	Descriptive statistic				Dendro-chron-ological prospective			Assessment of TRW-climate relationship																			Decadal Variations	Variations in TRW after year 2000			
								Correlation and regression analysis																							
	Vessel description		TRW					TRW and climate			TRW and precipitation (mm)				VD and Precipitation				TRW and maT(c)								1st Decade 1986-1996 2nd Decade 1997-2006 3rd Decade 2007-2016				
	MV (μ)	MV (mm²)	MV	SD	CV	CC	T	GLK	SI	MS	AC	R	r²	P. v	r	r²	Beta	Sig	R	r²	Beta	Sig	r	r²	Beta	Sig	Dec-ades	MV	In-crease	Decrease Increase	Decrease
Swat	101.883	13	1.00	0.20	20 %	0.38	2.9	0.66	0.330	0.349	0.025	0.201	4.04 %	0.046	-0.200*	4 %	-.0003	0.013	0.070	0.49 %	0.002	0.641	-.008	0.01 %	-0.004	0.820	1st	0.961		.016	-
																											2nd	1.063	.10		
																											3rd	1.123	.06		

and weak. Beta values reveals increase up to 1 unit in precipitation index may cause 0.002µm diameter increase; this may assume as high precipitation may result large diameter of the vessels. The results are an assumption due to short survey samples and may change if samples increase.

Conclusions and Recommendations

Walnut *(Juglans regia)* experiences high degree of annual variations due to climatic fluctuations. Dendrochronological potential of the species is moderately good enough, thus the chronology may be used as proxy of to study climatic variations. Climate-TRW relationship was found relatively significant showing moderate limitations of climate on the growth of the species in Swat area. Precipitation found as a strong growth limiting factor in comparison to temperature. Rainfall ratio was found on decline whereas rise in MAT, after the year 2000; this phenomenon affected the growth of the species and triggered fast radial growth in species. Moreover, increase in the diameter of the vessels was observed in the species more probably as a result of decline in precipitation in the area.

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Novelty Statement

In the shadow of shifting climate patterns, this study reveals a thoughtful investigation into the resilience of the walnut tree, shedding light on its adaptability potential within the dynamic environmental landscape of Swat, Pakistan.

This research investigates deep into the complex relationship between climate change and the anatomical characteristics of walnut trees, offering insights that could play a pivotal role in safeguarding one of the region's most cherished and economically significant tree species and biodiversity as well.

Author's Contribution

Concept and design, writing the first manuscript, literature review, data presentation, critical thinking, problem solving, collaboration, communication, editing and revisions by Khalid Hussain; compilation, tabulation, data analysis and proofreading by Tanvir Hussain; data collection, analysis and tabulation by Muhammad Inamullah Khan.

Conflict of interest

The authors have declared no conflict of interest.

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