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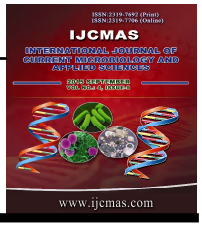

Original Research Article

Some Phenotypic Variations in the Population of White Olive

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Keywords

Olive, Genotype, Morphological analysis, Feature, Marker

A B S T R A C T

This study was performed during 2010-2014, to identify the variations within the population of Ullinj të Bardhë as well as their homonymy and synonymy. 13 ecotypes of this population were characterized with a morphological marker and were analysed with jmp software. The variables had considerable amplitude, but the main constituents of polymorphism were 6 elements which comprised 93.829% of the variability. The genotypes displayed deviations from the average of the standard population (Xm) as a result of environmental changes. In conclusion the genotyped derive from the same trunk (Ulli i bardhe - BT), but the reactions to climate availability have had changes under its influence. 10 genotypes are synonyms with UBT and result from the same variety called under different names. Cv Lundra (LU), Kçarr (KC) and Vajs Peqini (VP), represents polymorphism 13, 18 and 23% with a specific profile.

Introduction

The population of the Ulliri i Bardhë lies in the Mediterranean Adriatic region at a longitude 40-42 degrees and H 11-550 m *Hannachi, et al (2007) ; Ismaili et al., (2013*). Tirana, heart of the origin, displays a rich diversity (Olea Europaea L.), represented by cultivated as well as spontaneous forms, a real wealth which in fact is not well–known (*Belaj et al., (2011)* ; *Bartolini et al (1998)* . The programme of the Genetic Bank has important objectives for the recognition of the autochthonous diversity, study of the cultivars, ecotypes which are not widely known and preservation of the genetic wealth *Ismaili et al. (2013)*. Variability within one population is not only the result of genetic factors, but also of environmental influences especially the temperature, which has very big influence, *Arsel et al (994)*. Analysis which aim to find factors of standard deviance through variance of the relation genotype-environment are important in determining the phenotype *Bellini, (1993)* ; *Ismaili et al. (2012*), because our olive groves display genetic confusion, as different varieties are called under a common name, whereas the same variety is often named differently *Anguilillo et all (1999*). As of this phenomenon a lot of varieties are yet to be identified and described. Hence the object of this research is characterization of the ecotypes *Ulli i bardhe (UB*) based on endocarp and leaf as a stable important

marker stable in measuring their polymorphism *Padula et al., (2007) ; Cantini et al (1999); Pearce et al. (1967*).

Materials and Methods

Analysis was performed during 2010-2014 as part of the project of the genetic bank on the study of genetic diversity of the olive. The genetic material is made up of 13 genotypes which are faced with a problem of synonymy and homonymy within the population of the white olive. The genotypes have regional names as of cultivation area, as follows: *i Bardhi i Tiranes (BT), Kcarr (KC), Farka (FA), Bardhi Durrësit (BD), Bardhi Lezhës (BL), Bardhi Shkodrës (BS), Karen (KA), Bardhi Krujës (BK), i Bardhi Brarit (BB), Frëng (FR), Lundra (LU),Mixan (MI), and Vajs Peqini (VP).* The trees characteristics include 34 features: 20 quantitative and 19 qualitative, as of methodology cited by the International Olive Oil Council (Resgen, 1997), and by (UPOV, 1985). Every 100 leaves/year/genotype at the end of vegetation had the following main characteristics: height, width, surface, ratio L/l, form, symmetry etc. Every 100 fruit/tree/year at ripeness period: diameter (D), diameter (d), ratio D/d, average weight, form and symmetry. Every 100 endocarps the main measurements included: diameter (D), and(d), ratio D/d, average weight, numbers of grooves, form and symmetry, etc.*C.O.I., (1997) ; Idrissi et al. (2004*)

Statistical analysis

Descriptive analysis consisted of variation coefficient for the quantitative characters, standard deviation and Tukey test with a significant level of ($p<0.05$) per variance between genotypes and correlation with the environment. Moreover, the traits mean values were used to perform principal component (PCA) and cluster analyses using the Jmp software 2010. Finally, based on morphological similarity, cluster analysis was conducted on the Squared Euclidean Distance matrix with the Unweight Pair Group Method based on Arithmetic Averages (UPGMA). For the qualitative characters Nei and Shannon indices calculated the phenotypic frequencies and diversity per each character *Erfatpour, et al (2011)* ; *Hagidimitriou et al (2006*)

The diversity index Shannon-Weaver was provided via the formulae:

$$H = -\sum_{i=1}^{n} Pi\ (Pi)$$

where *Pi* is the proportion of genotypes in the *ith* class of an n-class character and *n* is the number of phenotypic classes for a character.

The diversity was also estimated by Nei a diversity index which is defined as: $H' \frac{2n}{2n-1}\ (1 - \sum P^2_i)$

where *Pi* refers to the frequency of descendants in each class for each character and *n* is the number of studied descendants

Results and Discussion

Variability in the population Ulliri i Bardhë: Phenotypic variation has brought about development after all, as it has caused heterogeneous production, different pathogenic periodicity and resistance. In *table 1*, Ulliri i Bardhë in the climate of Tirana (origin) has a value of (m+g), whereas in Peqin, Durrës, Elbasan, Krujë, Lezhe, Shkodër and Malësi e Madhe, the genotypes have displayed deviances (±) from (Xm). No matter what size the phenotypic variance of genotypes was, it was the product of genetic ($\delta.G^2$), and environmental causes ($\delta.E^2$) and was

calculated as of formula: ($\delta P^2 = \delta G^2 + \delta E^2$) F*ontanazza et al (1999) ; Gregoriou et al. (2006*). Between the two extremes of longitude comprising the environment Thermal constant(Te), Heliometric index (Ie), have considerable variance and variation coefficient is respectively Te=32%, St=21%, Ie=12%.

In Table 2, Only 9 of the quantitative characters are important (PC>0.35), because they have explained more than 93% of the variation (3 the leaf, 3 endocarp, 3 fruit, etc). Five characters in PC^1 explained 56.6% of the variation, respectively: (LL, LW, LA, FL, FSR, SG). PC^2 FW and FL have 25.2%. *PC_3, FW and SR had 11.9%* of variability. Performance of the three first PCs, in addition to qualitative characteristics facilitated the calculation of phenotypic variance in the olive population, *Poljuha et al. (2011*).

Quantitative traits: Descriptive analysis, in table 1 and 2, represented degree of deviation within the population per ($p < 0.001$) for quantitative characteristics.

Variance above cumulative values of 5 quantitative characteristics of the leaf presented amplitude of (308 - 444) frequencies, means 389,1 and variation (cv=9.25%), for *0.033**. Ulli i bardhe (BT) olive has leaves of short length (92%), small width (93%), and small size (92%), whereas 13 genotypes have variability and are classified in thre groups of certified variance. G_1, has 9 homogenous individuals including *ulli i bardhe* (BT). G_2, 3 individuals BL, BS, LU and G_3, is VP of variance level (28%) as opposed to G_1 and 11% of mean variance Tukey for level of significance *0.001* lsd. 1,11*.

Cumulative values for 8 quantitative characteristics of the endocarp extend with e frequency of (54-69.4), means 59.1 and cv=6.6%. Tukey test, for level 0.0001* and lsd1.112. As seen 10 genotypes do not change from *ulli i bardhe* (BT). They have endocarp of low weight (0.37), average number of grooves (9.1), which continue up to highest top (89%), have a elliptic shape of (80%), symmetric (78%), and slightly symmetric 12%.. Whereas two individuals Kcarr (KC) and Lundra (LU), display a variance level of 21.6%, (Symmetry, Shape, Weight).

Cumulative values for 5 quantitative characteristics of the fruit, comprise 8 genotypes including BT without proved changes (G_1). They have a symmetric shape (88%), are slightly symmetric (12%) and they are ovoide shape (90%). Ration between variances level to average variance is cv=3.3 %. G2 genotypes KA and LU have variation 12 and 13% and dominant position as opposed to G1, whereas 2 individuals from G3 (Kcarr & VP) have dominant variance (27.7%) from variance average. (G3>G2>G1, Fruit variance). Cumulative quantitative values are influenced by the environmental variations following longitude, while the ratio P/E does not display certified changes among the genotypes, *Fourati et al (2002*).

Qualitative characteristics of leave: Diversity's index *Table 2* shows the degree of diversity for the quantitative characteristics. G1 was considered for the leaf, where 11 individuals including BT, have leaf (SE) shape elliptic lanceloate 73.6%, with longitudinal curve, (Sel) shape elliptic 18.6%, close apical angle (AN) 93%, close basal angle (BN) (89%). Iponastic nervature (NF) nervation flat 74%. Genotypes KC and LU have certified changes compared to G1, cv=23%, cumulative qualitative characteristics of the leaf for 12 genotypes in G1, yield an

average of (88%), whereas in G2 both genotypes LU and VP have an average of 69%, in G3 one genotype (KC) with an average of 71%. The level of leaf variance of G2 and G3 is respectively 11 and 17% compared to average of variance. Leaves of BL and LU genotypes are more intense color (5) and leaves resistant cycloconium, Nei *et al (1979*).

Qualitative characteristics of endocarp: Shannon and Nei index, as a measure of morphological trait diversity were calculated for all qualitative parameters per 13 genotypes were presented in Table 2 and 3. Cumulative value represents diversity thus classifying the genotypes in three groups: ***first*** 10 genotypes including homogenous BT, with an average cumulative value 59: are asymmetric (SYas) 9.6%, (VP -22%), pointy sharp endocarp, (Ap) apex pointed-96%, round at the base (Br) base rounded 95%, supplied with mucron 77%, elliptic form She) 89%. (Shee), shape elip/elongated 9.6%. In G_2 1 genotype LU, cumulative average 69, and variance 13% form the average variances have pointy sharp endocarps (Ap)(59%) round form on top (39%), smooth surface (33%) and the number of grooves (11.1), ovoid stone (52%), a central maximum diameter, a rough surface and a uniform grooves. G3 has genotypes; VP and KC, which have cumulative average 84 with a variance level 12% compared to average variance. Variance derived from the differences in form and asymmetry.

Figure 4, Dendrogram of Shanon and Nei index values showed that correlation strength R^2 is 0,92 which simultaneously shows phenotype variance. These indices showed similar trends in phenotypic diversity. *Figure 4*, displays high coefficient of correlation between two indices of diversity and R^2 resulted 0.91. The highest values of index diversity were identified in stone characteristics for BT and stone of LU. Average Shannon index was equivalent to 0.54 and fluctuated from 0.43 to 0.59. The highest diversity index values were noted on stone weight (SW) and leaf. Average Shannon and Nei diversity index was respectively to 0.55 and 0.36, ‘

Finally, as of figure 3, phenotypic differences were identified in the characteristics of endocarp and leaf. General evaluation of genotypes displayed similarity level of 83.5%, whereas 16.5% is obviously the variance of the environmental and agro-biological influence. Values of the three first PC showed the degree of similarity and presented a trunk with two primary and two secondary branches. This is obviously the result of the deviance and phenotype polymorphism caused by the environment that BT has undergone pursuant to the period of emerging from the origin. But as of *dendogram-5*, 7 genotypes have great similarity and constitute morphologic synonymy, whereas G_2 comprises Lundr and Kcarr with variability 29%, which comprise two specific ecotype. Dominant characters of the variance were leaf symmetry, form and resistance to cycloconium oleaginum. BLE in the first group varies because there are 38% highest resistance.

Regression analysis of relations for cumulative quantitative characteristics (y) showed the trend for changes in relation to any change of effective temperature (x) with value, r^2= 0.838 shows that influence of tempreature in the values of phenotype change is 16.2%. Tested relation (xy), has defined the sense towards change between the two characteristics (x) and (y) and regression presented the values of phenotype changes (y) per unit of the factor (x) (environment). The relations were of negative or positive sense per each analysed couple.

Table.1 Descriptive statistical analysis of 20 quantitative morphometric traits evaluated for 13 ecotypes in the population of BT Cv

Genotyp	LL	LW	LR (L/l)	LA mm²	FW	FL	FWI	FR	SW	SG	FSR.	SL	SWI	SR
BTIR	44	9	4.8	328	235	21.5	14.6	1.4	34	8.3	5.9	13.9	6.4	2.2
KCAR	43	9.3	4.6	353	290	22.4	15.2	1.4	36	9.5	7.0	14.2	7.1	2
FARK	44	9	4.8	323	233	22.3	14.8	1.5	38	7.4	5.1	13.6	7	1.9
BDUR	45	9	5	318	227	21.5	14.6	1.5	34	9.7	5.6	13.9	6.4	2.2
BLE	47	11	4.3	382	237	21.2	14.4	1.5	35	8.9	5.7	13.5	6.5	2.2
BSHK	52	11	4.7	341	231	19.5	14.1	1.4	35	10.1	5.6	13.4	7.1	1.9
KAREN	43	8.9	4.8	313	223	18.4	13.1	1.4	34	8	5.5	12.6	6.2	2.0
BKRUJ	50	10	5	379	252	21.2	13.6	1.5	35	10.2	6.2	11.8	5.6	2.1
BRAR	45	9	5	329	245	19.1	13	1.5	38	8.2	5.4	13.5	6.2	2.1
LUNDR	40	8.3	4.8	300	224	19.8	13.3	1.5	45	11.1	4	15.3	6.9	2.2
FRENG	51	11	4.6	346	228	19.7	14.4	1.5	38	8.6	5	14.5	6.6	2.2
MIX	41	9	4.5	335	232	19.8	13.7	1.4	38	10.4	5.1	13.3	6.2	2.1
VPEQ	38	8	4.5	258	214	23.2	16	1.45	40	8.6	4.3	13.7	6.5	2.1
Min	38	8	4.3	258	214	19.1	13	1.4	34	7.4	4	11.8	5.6	1.9
Max	52	11	5	382	290	23.2	16	1.5	45	11.1	7.0	14.5	7.1	2.2
Mean	44.8	9.42	4.7	330	236	207	142	1.45	37	9.15	5.42	13.9	6.5	2.04
StdDev	4.18	1.03	0.24	33.3	18.4	14.2	8.61	0.05	3.08	1.02	0.74	0.85	0.42	0.11
StdErr	0.66	0.16	0.03	5.34	2.95	2.31	1.37	0.00	0.49	0.23	0.12	0.13	0.06	0.01
CV	9.3	10.9	5.1	10.1	7.8	6.9	6.0	3.4	8.3	11.1	13.6	6.1	6.4	5.3

LL: leaf length (cm), LWI: leaf width (cm), LA: leaf area (mm^2), LR: leaf (length/width) ratio, FW: fruit weight (g), FL: fruit length (mm), FWI: fruit width (mm), FR: fruit (length/width) ratio, SW: stone weight (g), SL: stone length (mm), SWI: stone width (mm), SR: stone (length/width) ratio, SG: number of grooves, FSR: fruit flesh to stone ratio. SD: standard deviation, CV: variation coefficient ; CL-cum leaf, CF- Cum Fruit. Sig Level: significance level, ***significant at 1‰ level.

Table.2 Descriptive statistical analysis of the qualitative traits of leaf and stone, evaluated for 13 olive tree in populations of BT

Genotyp	SL	SE	Sel	NF	NH	NE	AN	BN	SHs	SHo	SHe	SHe	SYs	SYa	Ap	Ar	Bp	Br	Bt
BTiran	0	77	23	77	0	23	91	86	0	0	90	10	90	10	100	0	7	93	0
Kçarr	13	60	27	54	0	46	88	88	0	0	94	6	95	5	100	0	1	99	0
Farka	2	75	23	88	2	10	100	86	0	0	92	8	94	6	100	0	7	93	0
BDurr	2	76	22	68	0	32	90	90	0	0	91	9	93	7	100	0	5	95	0
BLezh	3	83	14	90	0	10	93	93	0	0	93	7	91	9	100	0	0	100	0
BShk	5	75	20	65	0	35	89	90	0	0	94	6	95	5	100	0	9	91	0
Karen	6	78	16	60	0	40	99	94	0	0	92	8	94	6	100	0	11	89	0
BKruj	2	70	28	67	0	33	88	91	0	0	91	9	90	10	100	0	5	95	0
BBrar	3	75	22	76	0	24	90	86	0	0	94	6	92	8	100	0	4	96	0
Lundra	43	57	0	90	0	10	99	98	0	52	48	4	88	12	59	41	1	99	0
Freng	5	77	18	78	0	22	94	87	0	0	94	6	88	12	100	0	1	99	0
Mixan	3	80	17	73	0	27	95	88	0	0	96	4	88	12	100	0	2	98	0
Vajs Peqin	12	75	13	79	0	21	98	90	0	0	85	15	78	22	91	9	10	90	0

SL-shape lanceloate**; SE**-shape elliptic lanceloate, **Sel**-shape elliptic; NF-nervation flat, NH-nervation hyponastic, NE- nervation epinastic, AN- apex narrow, AS-apex spread, BN-Base narrow, BS-base spread; **SHs**-shape spherical**; SHo**-shape ovoid**; She**-Shape elliptic; **SHee**-shape elip/elongated; **SYs**- symmetric; **SYas**-asymetric; **Ap**-apex pointed; **Ar**- Apex rounded**; Bp**- base pointed**; Br**- base rounded**; Bt**- base tapered.; **SHs**-shape spherical**; SHo**-shape ovoid**; She**-Shape elliptic; **SHee**-shape elip/elongated; **SYs**- symmetric; **SYas**-asymetric; **Ap**-apex pointed; **Ar**- Apex rounded**; Bp**- base pointed**; Br**- base rounded**; Bt**- base tapered

Table.3 Evaluation of variances, variances accumulated coefficient Eigen value, (autovectors) for the first three principal components, evaluated with 20 quantitative traits

	PC1	PC2	PC3
Eigen value %	**56.666**	**25.210**	**11.953**
Cum.Variation %	**56.666**	**81.876**	**93.829**
L.L	0.37415	-0.34138	-0.10999
LW	0.37785	-0.33793	-0.04906
L.A	0.42717	-0.07192	0.27898
C.L	0.33395	-0.12568	0.26052
F.W	0.26974	0.45097	0.40067
FSR	0.42221	0.32604	-0.01375
F.L	-0.02475	0.55821	0.01960
C.S	-0.31825	-0.12726	0.31340
S.R	-0.20149	-0.18872	0.71260
L.R	0.21862	-0.03125	0.15629
SG	0.36822	0.15477	0.14945
C.F	0.03086	-0.05779	-0.31759
F.W.I	-0.15030	0.23389	0.24436
F.R	-0.26916	-0.04848	0.26666
S.W.I	-0.13802	-0.55257	0.25938

Figure.1 The hearth of White olive and the geographical position of 13 olive genotipe, in situ

Figure.2 Photo of stone in the white olive population

Figure.3 Plot illustrative for links that exist between the 13 olive genotype based on quantitative morphological traits and their distribution

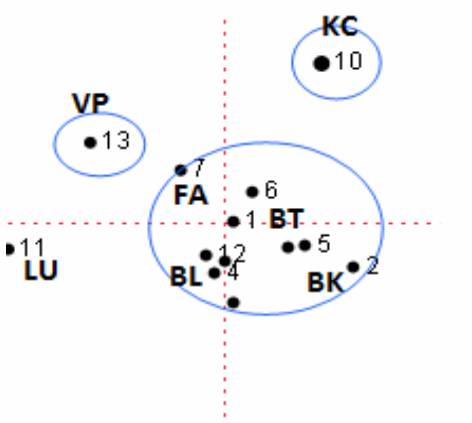


Figure.4 The correlation between Shannon and Nei indices analysis of 13 genotypes of olive on 19 qualitative traits

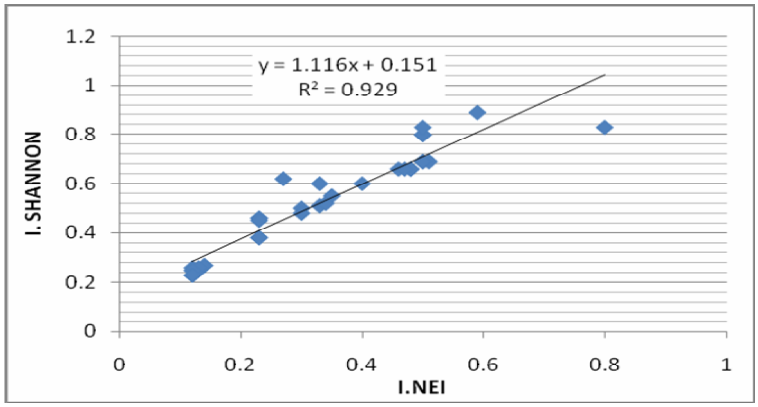
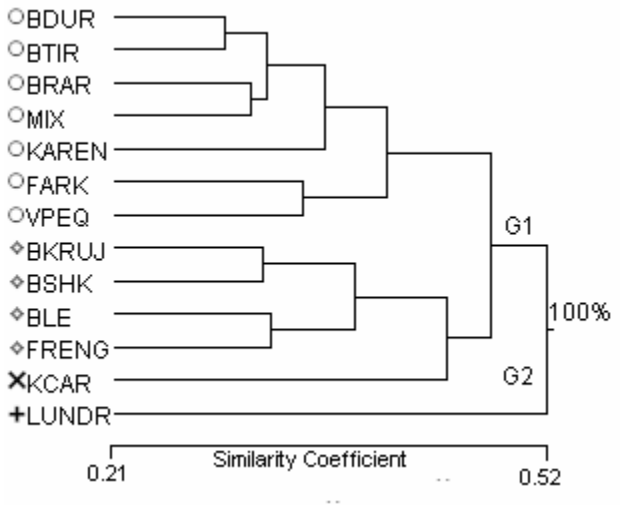


Figure.5 UPGMA dendrogram based on Spearman'p coefficient illustrating the genetic similarities and distance among olive cultivars



Phenotypic variation in the population of UB has not been considerable and the degree of changed phenotype was in correlation with the general variance of the environment.

Fruit colour, leaf, form and symmetry, endocarp and the type of grooves, connection of two poles, resistance to fungi etc, were the most important phenotypic variations.

To sum up phenotypic identification in the population of Ullinj te Bardhe are attributed to two sources causing of the variation: genetic plasticity and environmental influence. 10 ecotypes resulted of the same variety which has been named differently as of cultivation area. BL cv, Express variation for the resistance to Cycloconium. Lundra, VP and Kcarr are with varietal particular profile.

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