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Complex Segregation Analysis of Body Height, Weight and BMI in Pedigree Data from Middle Dalmatia, Croatia

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A B S T R A C T

It has recently been reported that the mode of inheritance of body height, weight and BMI in five ethnically and geographically different populations can be described in terms of a major gene (MG) model[1]. Here, using the pedigree sample from the island populations of Middle Dalmatia, Croatia (1,312 observed individuals in 462 pedigrees), the evidence is presented that supports the above findings. By applying the usual transmission probability tests, the hypothesis has been accepted that a significant part of the variation of each one of those three basic morphological traits can be attributed to the effect of a putative large-effect gene. The effect of a putative MG is responsible for 39–50% of age and sex adjusted trait's variation and for 34–48% of the total (non age-adjusted) variation of height, weight and BMI.

Key words: *complex segregation analysis, body height, body weight, BMI, Croatia*

Introduction

Attempts at defining the genetic basis of fundamental characteristics of human morphology – such as body height and weight – date back to the very beginning of the quantitative genetics. Existence of a genetic component in variation of the body height, as well as body weight and body mass index (BMI) has been documented by numerous family and twin studies[2–7].

Significance of genetic control of morphological traits has also been indicated

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by the population studies. For example, the analyses carried out on the population groups of Middle Dalmatia, Croatia, based, among others, on the distance analysis of various measures of biological (polygenic and monogenic), socio-cultural and bio-cultural traits, showed that subpopulation heterogeneity of morphologic features follows the genetic pattern expected by known historical migrational movements[8–12]. Environmental factors are also very influential, in particular, a strong negative influence of socio-economic deprivation in the growth and development period was well established[13].

Researches conducted in the fields of genetic epidemiology and molecular genetics over the last 10 years – primarily oriented on obesity-connected traits – clearly documented that obesity as well as susceptibility for gaining or loosing weight are largely determined by gene effects. Recent heritability estimates for BMI are dominantly reported to be within the limits of 50–80%[14–18]. A possible existence of discriminative one-gene effects in the development of morphological features has also been investigated and a major gene (MG) effect on BMI has been documented in several studies[1,19–25].

The inheritance of BMI, surely being far from a simple monogenic mode, is influenced by various other factors that modify that complex phenotype. Age has an especially significant influence on BMI variation (therefore on it's heritability estimates as well), as well as the gender has its genotype/environment specific effects[6,23,26–28]. However, there is also increasing evidence on the possibly significant role of population differences in gene frequency[19,21,29].

Evidences for oligogenic inheritance of the two other traits under study, the body height and weight, are not so definite. Recently, Perola et al. (2001)[30] reported 2 chromosomal regions showing linkage with height. Ginsburg and co-workers (1998)[1] reported results of segregation analysis of body height, weight and BMI performed on pedigree samples collected from five ethnically and geographically different populations (from Kirghizstan, Turkmenia, Chuvashia, Israel and Mexico). By the usual transmission probability tests, the MG mode of inheritance was not rejected for each of these three studied traits on all 5 populations. Since we find it very important to test whether the results found by Ginsburg et al. (1998)[1] can be taken as a general phenomenon, in this study, undertaken by the implementation of the same methodology (i.e. program package MAN), the segregation analysis of body height, weight and BMI has been performed using the pedigree data from the islands' population of Middle Dalmatia, Croatia.

Material and Methods

Original sample

The data on body height, weight, and body mass index (BMI), as well as the information regarding the family relations used in this study, are a subset of the extensive material collected from 1978 to 1987, from the population of islands of Brač, Hvar, Korčula, and Pelješac peninsula, by the research team of the Institute for Anthropological Research, Zagreb, Croatia, lead by Professor P. Rudan. The data have been gathered from randomly sampled individuals, encompassing from 6.2% (Korčula) to 10.7% (Brač) of the total population of those islands/peninsula. Middle Dalmatian islands of Brač, Hvar, Korčula, and Pelješac peninsula are neighboring islands/peninsula that occupy a rather small area and their inhabitants share the same environmental conditions (climate, professions, economy, culture, health service, life style). Modern population of those islands/peninsula is composed partially from the ancient Croatian population (which from

the 7th century gradually slavicized the remains of the romanized Ilirian population) and partially from the new Croatian population which intensively migrated to this area from the east during the Turkish wars, from the end of 15th to the end of 17th century. Various ethno-historical and biological properties of that population have been intensively investigated and the relevant data can be found elsewhere[8–12,31–43].

Body height and weight measures were taken according to IBP recommended technique using standard anthropometric instruments (Weiner and Lourie, 1981)[44]. Body height was measured using a Martin metal anthropometer to the nearest 0.1 cm and body weight was recorded with a portable scale precise to 0.1 kg. Body mass index (BMI) was calculated as body weight (kg) / body height $(m)^2$.

Pedigree data

The number of subjects included in this study (1,312 examinees, 610 males and 702 females, aged 17 to 87) was determined by the coincidence that two (or more) participants of the original random sample are the members of the same family. The sample consisted of 462 two and three-generation pedigrees and their size was distributed as follows: 271 pedigrees having 2 observed individuals, 108 having 3 individuals, 72 having from 4 to 6 individuals and 11 families having from 7 to 18 individuals each.

Two factors caused certain specificity in preparing the pedigree data for segregation analysis. The first one was the significant difference between some of the compared 4 sub-samples (i.e. samples from the islands of Brač, Hvar and Korčula and Pelješac peninsula). The second factor was connected with the segregation analysis aim, which is to test various hypotheses about the mode of inheritance of body height, weight and BMI in *normal* population. Defining the »norm«, it was assumed that 1) The trait values that differ from the population mean more than 3SD are considered outlying; and 2) The difference between the trait values in parent and offspring or in any sibling pair (if they are truly related) also should not exceed 3 standard deviations. Accordingly, the pedigree data were prepared in two successive stages as follows:

1. Body height and weight measures were adjusted for sex and age and standardized within each of the sub-populations (4 islands/peninsula) separately. In cases when the difference between standardized values of height and weight in an individual exceeded 3, his/her measure was excluded from the sample (see an example in Figure 1).

2. Next, the standardized data from each island/peninsula were pooled together forming the population pedigree data and the second stage of the data clearing was performed. If the difference between standardized trait values in parent-offspring or in sibling pair exceeded 3, then one of the pair was excluded from the analyzed sample (see an example in Figure 2).

Segregation analysis

Segregation analysis of pedigree data has been performed by means of the program package MAN developed by Ginsburg (1997)[45]. The major gene (MG) model of quantitative trait inheritance is described by the following parameters:

p – is the population frequency of the first of two MG alleles (A_1 and A_2);

μ_g – is the average trait value (genotypic value) in all individuals having genotype g; g = 1, 2 and 3, corresponding to genotypes A_1A_1, A_1A_2 and A_2A_2, respectively.

σ^2_g – is the trait variance in individuals having the same MG genotype g. It estimates the residual trait variation, i.e. the variation due to effects

of all possible environmental factors and potential minor genes;

ρ, β, ε – are the partial correlations between residuals in spouses, parents and offspring, and in sibs, respectively, estimating all non- MG effects causing co-segregation in relatives; partial correlation between residuals in any pair of relatives not-belonging to the same nuclear pedigree was assumed equal to zero;

Note that all the above given parameters correspond to those used in the program package S.A.G.E. (Elston, 1995)[46] and PAP (Hasstedt, 1994)[47], taking into account that the partial residual correlations are used here instead of the pairwise ones.

The MG hypothesis was tested by means of two transmission probability tests (Elston and Stewart, 1971)[48], namely, $\chi^2_3 = 2[LH(\hat{\tau}) - LH(\tau_0)]$ and $\chi^2_2 = 2[LH(\hat{\tau}) - LH(\bar{\tau})]$, where $LH(\tau)$ is the maximum log-likelihood value obtained with transmission probability $\tau_g = Pr(A_1|g)$; τ_0 denotes Mendelian transmission probabilities, 1.0, 0.5 and 0.0 for the parent's genotype g = 1, 2 and 3, respectively; $\hat{\tau}$ denotes the maximum likelihood estimates of these probabilities (general model), and $\bar{\tau}$ is the maximal likelihood estimate for transmission probabilities constrained to be equal. The MG model of the trait inheritance is accepted if 1) χ^2_2 exceeds the critical value corresponding to df = 2 and the *a priori* established type I error α = 0.01 (the hypothetical independence of offspring's MG genotype from the genotypes of his/her parents is rejected), and 2) concurrently, χ^2_3 does not exceed the critical value corresponding to df = 3 and type I error α = 0.05 (the hypothesis of Mendelian transmission probabilities is accepted).

No ascertainment correction of the pedigree likelihood was made because the pedigrees were collected randomly.

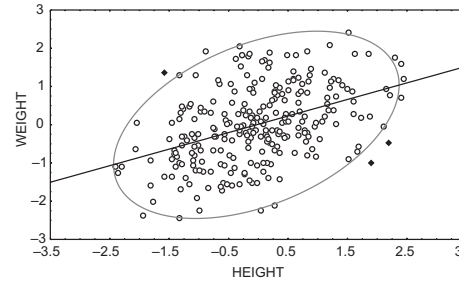

Fig. 1. The 1st step of pedigree data preparation: Scatterplot shows height (x) and weight (y) of females from the island of Brač (data are age-adjusted and standardized within every island separately). Excluded individuals (3 cases) by criterion of difference between weight and height exceeding 3 standard deviations are denoted by black rhombs. The ellipse is marking the area where the values will fall within with 95% probability, with the assumption that the two variables follow the bivariate normal distribution.

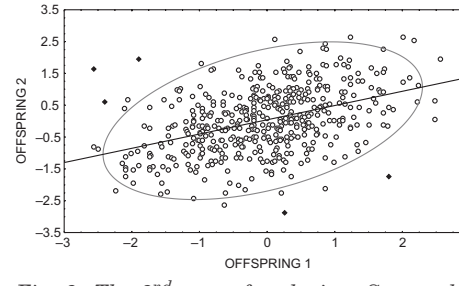

Fig. 2. The 2nd step of exclusion: Scatterplot shows body height in sibling pairs from all four sub-populations (data are age adjusted and standardized within every island separately). Excluded individuals (5 cases) by criterion of difference between values in first (x) and second (y) offspring exceeding 3 standard deviations are denoted by black rhombs.

Barać and coworkers (1999)[43] have found no tendency of preferential selection of mates that are similar/nonsimilar in physical characteristics in the population of the island of Brač (as well as for the islands of Hvar, Korčula and Pelješac peninsula (data not published, yet). They

reported that two most important elements for choosing the mate in this population are: a) geographical closeness (i.e. the mates are coming from the same village) and b) age difference of mates[43]. Accordingly, no assortative mating effect was explicitly included in the MG model and tested.

Additional characteristics of the tested model evaluating its fit to the analyzed pedigree data were: H^2 – the trait heritability, evaluating the proportion of phenotypic variance attributable to the putative MG effect; D^2 – the proportion of the trait variation attributable to both the MG effect *and* the non-MG (multifactorial) effects described by correlations ρ, β and ε; and the correlation $R(x,\tilde{x})$ – measuring the prediction ability of the chosen model, where x denotes the measured trait value in an individual and $\tilde{x}$ is the trait value as predicted by the chosen MG model on the basis of trait values of the individual's relatives[1,45].

For each studied trait, the most parsimonious (MP) model was obtained using the algorithm proposed by Ginsburg et al. (2001)[49]. In the process of MP model construction, all possible constraints of the parameters are tested stage by stage and if not rejected they are incorporated in the model up to the limit when any further parameter constraint is statistically rejected. During this process, the hypotheses about the additive, dominant and recessive MG control are tested, as well as

TABLE 1
DESCRIPTIVE STATISTICS (N, X, SD) OF AGE, BODY HEIGHT, WEIGHT AND BMI IN FOUR SUB-SAMPLES (ISLANDS/PENINSULA) AND IN TOTAL PEDIGREE SAMPLE FROM MIDDLE DALMATIA, CROATIA

Population	Trait	Males			Females		
		N	X	SD	N	X	SD
Brač	Age	222	48.56 ^{H,K,P}	15.11	274	48.91 ^{K}	15.70
	Height	221	1.745 ^{K,P}	0.073	272	1.617 ^{P}	0.066
	Weight	219	83.02 ^{H}	11.73	266	73.55 ^{H,K}	11.43
	BMI	218	27.22 ^{H,P}	3.29	264	28.16 ^{H,K,P}	4.29
Hvar	Age	111	44.87 ^{B}	14.30	118	48.41 ^{K}	11.26
	Height	109	1.752 ^{P}	0.062	115	1.619 ^{P}	0.056
	Weight	111	75.97 ^{B,K,P}	9.62	116	66.35 ^{B,K,P}	9.86
	BMI	111	24.77 ^{B,K,P}	2.82	116	25.38 ^{B,K,P}	3.77
Korčula	Age	126	42.74 ^{B}	11.86	159	44.57 ^{B,H,P}	12.28
	Height	125	1.766 ^{B,P}	0.068	158	1.626	0.048
	Weight	126	82.74 ^{H}	12.39	159	69.82 ^{B,H,P}	9.48
	BMI	125	26.52 ^{H}	3.51	158	26.43 ^{B,H}	3.56
Pelješac	Age	158	45.20 ^{B}	15.62	158	50.23 ^{K}	14.58
	Height	155	1.785 ^{B,H,K}	0.067	157	1.638 ^{B,H}	0.061
	Weight	156	83.01 ^{H}	12.30	158	72.43 ^{H,K}	11.17
	BMI	155	26.02 ^{B,H}	3.58	158	26.99 ^{B,H}	4.05
Total	Age	622	45.84	14.63	709	48.15	14.17
	Height	610	1.761	0.070	702	1.624	0.060
	Weight	612	81.68	11.95	699	71.25	10.99
	BMI	609	26.32	3.44	696	27.04	4.11

the hypothesis of no-MG control on the trait variability.

Results

Pedigree data

The descriptive statistics of age, body height, weight and body mass index (BMI) in 4 sub-populations (i.e. inhabitants of Brač, Hvar, Korčula and Pelješac) and in total sample, for males and females, is presented on Table 1. The differences (according to *t*-test) in above traits between 4 sub-populations is also shown on this table by superscript letters indicating island/peninsula of the sub-sample to which the trait in question proved to be significantly different.

The samples from the 4 neighboring islands/peninsula show few consistent differences: the females from Korčula are younger, and the males from Brač are older than others, both the males and females from Pelješac are taller then others, and the males from Hvar have lower weight and BMI values than the males from other sub-samples. All other differences do not follow any clear pattern (including the geographic one) but they just show that every sub-sample has its speci-

TABLE 2
INITIAL SAMPLE SIZES (4 SUB-SAMPLES AND TOTAL SAMPLE) AND SAMPLE SIZES AFTER 1ST AND AFTER 2ND (FINAL) STAGE OF PREPARATION OF THE PEDIGREE DATA FOR SEGREGATION ANALYSIS

Population	Stage	Height		Weight		BMI	
		No. of pedigrees	No. of individuals	No. of pedigrees	No. of individuals	No. of pedigrees	No. of individuals
Brač	Initial	138	493	138	485	138	482
	1st stage	138 0.000	488 0.010	138 0.000	475 0.021	137 0.007	471 0.023
	2nd stage	137 0.007	473 0.041	137 0.007	457 0.057	135 0.022	453 0.060
Hvar	Initial	90	224	92	227	90	224
	1st stage	89 0.011	219 0.022	87 0.054	215 0.053	87 0.033	215 0.040
	2nd stage	85 0.055	209 0.067	83 0.097	205 0.096	83 0.067	205 0.078
Korčula	Initial	122	283	123	285	122	283
	1st stage	121 0.008	281 0.007	93 0.081	213 0.253	92 0.246	211 0.254
	2nd stage	118 0.033	272 0.039	87 0.293	198 0.305	87 0.287	199 0.297
Pelješac	Initial	112	312	113	314	112	312
	1st stage	110 0.018	307 0.016	108 0.044	303 0.035	108 0.037	300 0.040
	2nd stage	108 0.036	300 0.039	106 0.072	294 0.064	106 0.053	291 0.067
Total	Initial	462	1312	466	1311	462	1295
	1st stage	458 0.009	1295 0.013	426 0.107	1206 0.080	424 0.082	1197 0.076
	2nd stage	448 0.030	1254 0.044	413 0.111	1154 0.119	410 0.112	1148 0.114

ficity, which provided us with an argument for performing age and sex adjustment of data on the sub-populational level.

Table 2 gives the size of each sub-sample and the total sample size after the two previously described stages of the data exclusion. Note that if the excluded individual was a member of a pedigree containing only two observed relatives or he/she was the only offspring in the family, then the entire pedigree was excluded from segregation analysis. In particular, the large percentage of excluded individuals in Korčula sub-sample (for weight and BMI) was caused by this very reason. Namely, most pedigrees collected from this island were nuclear consisting of only two first-degree relatives and therefore, virtually each exclusions of an individual led to the exclusion of the entire pedigree.

TABLE 3
CORRELATIONS BETWEEN BODY HEIGHT, BODY WEIGHT, BMI AND AGE IN TOTAL PEDIGREE SAMPLE

Trait	Weight	BMI	Age
Height	0.476***	–0.063*	–0.315***
Weight	–	0.842***	0.085**
BMI	–	–	0.347***

* p≤0.05; ** p≤0.01; *** p≤0.001

Inter-correlation of body height, weight, BMI and age for the total sample is presented in Table 3. Expectedly, BMI values showed strong positive correlation with age (e.g. Smolej-Narančić, 1999)[50]. However, the same age trend was not detected for body weight, as the result of a higher stature of younger individuals as the consequence of the combined effects of the aging process and secular trend present in this population[50,51]. The proportions of the variance attributable to the age effects was different for each trait and for each sub-sample ranging from R^2 = 0.003 for body weight in Hvar to R^2 = 0.188 for BMI in Pelješac sub-sample. For the total sample, the proportions of the variance attributable for the age variability were: R^2_{ht} = 0.099 for body height, R^2_{wt} = 0.007 for body weight and R^2_{BMI} = 0.120 for BMI. Table 4 presents family correlations for each trait after its adjustment for sex and age effects. The correlations have been calculated as pairwise for the parent-parent and parent-offspring pairs and as the intraclass correlation for siblings.

Segregation analysis of body height

The segregation analysis of body height was performed on pedigree sample consisting of 448 pedigrees (1,254 observed individuals) and the results are given in

TABLE 4
FAMILY CORRELATIONS FOR BODY HEIGHT, WEIGHT AND BMI. THE NUMBERS OF FAMILY PAIRS ARE SHOWN IN PARENTHESES. FOR INTRACLASS CORRELATIONS IN SIBLINGS, TWO DEGREES OF FREEDOM ARE SHOWN

Trait	Family correlations		
	Spouses	Parent-offspring	Siblings
Height	0.178 ns (121)	0.471*** (501)	0.468*** (327, 383)
Weight	0.177 ns (107)	0.343*** (462)	0.355*** (292, 342)
BMI	0.111 ns (106)	0.217*** (452)	0.319*** (290, 340)

ns p > 0.05; * p≤0.05; ** p≤0.01; *** p≤0.001

Table 5. The first three columns of the Table 5 present estimates of parameters and maximal log-likelihood value for the general MG model (column 1), for its version with Mendelian transmission probabilities (column 2) and the version with equal τ's (column 3). As seen from the comparison of the models presented in columns 2 and 3 with the first one, the transmission probability tests permit to accept the MG model of inheritance of body height: $\chi^2_3 = 3.36$ ($p > 0.05$) and $\chi^2_2 = 41.88$ ($p < 0.01$). The last three columns present the most parsimonious MG model (column 5), and its versions with arbitrary and equal τ's (columns 4 and 6, respectively). As seen from Table 5, the mode of inheritance of body height can be satisfactorily described by the MG model with codominant MG effect on the trait level, no differences among residual variances in three genotypes, and zero correlation between residuals in spouses. Bearing in mind the algorithm used here for construction of the most parsimonious model[49], it should be noted that the hypotheses of the dominant and recessive MG control of the trait, the hypothesis of no MG effect on the residual variance, as well as the hypothesis that no MG takes place in the trait control, have been tested and if statistically rejected their results are not shown in tables.

The MG effect is responsible for $H^2 = 0.485$ of body height variation (sex and

TABLE 5
SEGREGATION ANALYSIS OF BODY HEIGHT (STANDARDIZED VALUES) IN PEDIGREE SAMPLE FROM MIDDLE DALMATIA, CROATIA

Parameter	General models			Most parsimonious models		
	General 1	Mendelian 2	Equal τ's 3	Arbitrary 4	Mendelian 5	Equal τ's 6
p	0.778	0.727	1.000+	0.568	0.521±0.067	0.112
μ_1	–0.564	–0.611	–0.028	–0.887	–0.928±0.136	–0.323
μ_2	0.544	0.503	–0.227	0.078#	0.041#	–0.154#
μ_3	1.648	1.603	0.079	1.043	1.011±0.148	0.014
σ^2_1	0.614	0.584	1.016	0.506	0.499±0.050	0.938
σ^2_2	0.382	0.365	1.090	0.506!	0.499!	0.938!
σ^2_3	0.358	0.360	0.851	0.506!	0.499!	0.938!
ρ	0.078	0.079	0.092	[0.000]	[0.000]	[0.000]
β	0.221	0.217	0.265	0.245	0.243±0.048	0.274
ε	0.171	0.173	0.153	0.155	0.156±0.032	0.143
τ_1	1.000+	[1.000]	0.121	1.000+	[1.000]	0.000+
τ_2	0.384	[0.500]	0.121!	0.427	[0.500]	0.000!
τ_3	0.000+	[0.000]	0.121!	0.000+	[0.000]	0.000!
LogLH	–1680.72	–1682.40	–1701.66	–1683.36	–1684.07	–1703.45
χ^2	–	3.36 ns (1; df=3)	41.88 ** (1; df=2)	5.28 ns (1; df=4)	1.42 ns (4; df=3)	40.18 ** (4; df=2)

ns $p > 0.05$; * $p < 0.05$; ** $p < 0.01$; (N) number indicating the comparative column; [ ] parameter is fixed to shown value; ! parameter is constrained to be equal to the parameter above in the Table; # model is codominant; + parameter estimate achieved its limit. For parameter definitions see Material and Methods section.

age adjusted), while $D^2 = 0.570$ of its variation can be explained by all (genetic and environmental) family effects included in the most parsimonious MG model. For the trait measurements not adjusted for age, this means: $H^2_{ht} = H^2(1 - R^2_{ht}) = 0.4846\ (1 - 0.1492) = 0.4123$, and $D^2_{ht} = D^2(1 - R^2_{ht}) = 0.4852$. For body height, the correlation between the measured values (adjusted for age and sex) and the values as predicted by its most parsimonious model, $R(x,\tilde{x})$, was 0.521, $n = 957$, for offspring; 0.475, $n = 427$, for parents; and 0.506, $n = 1384$, for the total sample. The last number exceeds the number of individuals in the sample ($n = 1{,}254$) because in complex three-generation pedigrees, some individuals are offspring in one nuclear pedigree and, at the same time, they are parents in another nuclear pedigree.

Segregation analysis of body weight

Segregation analysis of body weight was performed on 413 pedigrees (1,154 observed individuals) and the results are shown in Table 6, which structure is the same as that of Table 5. As seen, the MG mode of the weight inheritance can be accepted when the general MG model is tested, as well as for it's most parsimonious version. The hypothesis about codominant MG effect was rejected, as well as that of dominant and recessive MG control (not shown) and that with no MG in the trait control at all. The accepted most parsimonious model showed dominant effect on residual variances ($\sigma^2_2 = \sigma^2_3$). The MG heritability was found to be $H^2 = 0.500$ and the proportion of the adjusted trait variance, explained by the most parsimonious model (D^2) was equal 0.544,

TABLE 6
SEGREGATION ANALYSIS OF BODY WEIGHT (STANDARDIZED VALUES) IN PEDIGREE SAMPLE FROM MIDDLE DALMATIA, CROATIA

Parameter	General models			Most parsimonious models		
	General 1	Mendelian 2	Equal τ's 3	Arbitrary 4	Mendelian 5	Equal τ's 6
p	0.698	0.729	0.444	0.581	0.623±0.043	0.406
μ_1	–0.523	–0.505	0.044	–0.767	–0.713±0.107	0.101
μ_2	0.351	0.332	–0.385	0.152	0.149±0.104	–0.403
μ_3	1.785	1.786	0.859	1.418	1.416±0.141	0.740
σ^2_1	0.682	0.704	0.903	0.586	0.638±0.097	0.821
σ^2_2	0.450	0.457	0.795	0.369	0.402±0.059	0.788
σ^2_3	0.204	0.202	0.700	0.369!	0.402!	0.788!
ρ	0.120	0.112	0.060	0.199	0.181±0.139	0.155
β	0.188	0.181	0.301	0.168	0.149±0.060	0.232
ε	0.185	0.178	0.124	0.198	0.185±0.043	0.175
τ_1	1.000+	[1.000]	0.838	1.000+	[1.000]	0.702
τ_2	0.591	[0.500]	0.838!	0.549	[0.500]	0.702!
τ_3	0.000+	[0.000]	0.838!	0.109	[0.000]	0.702!
LogLH	–1580.16	–1581.23	–1591.47	–1581.22	–1582.93	–1591.18
χ^2	–	2.14 ns (1; df=3)	22.62 ** (1; df=2)	2.12 ns (1; df=1)	3.42 ns (4; df=3)	19.92 ** (4; df=2)

ns $p > 0.05$; * $p < 0.05$; ** $p < 0.01$; (N) = number indicating the comparative column; [ ] parameter is fixed to shown value; ! parameter is constrained to be equal to the parameter above in the Table; # model is codominant; + parameter estimate achieved its limit.

which means proportions H^2_{wt} = 0.476 and D^2_{wt} = 0.518, respectively for the trait measurements not adjusted for age. The prediction ability of this model was evaluated by $R(x,\tilde{x})$ = 0.404, n = 883, for offspring; 0.346, n = 395, for parents; and 0.385, n = 1278 for the total analyzed sample.

Segregation analysis of BMI

Segregation analysis of body mass index (BMI) was performed on 410 pedigrees (1,148 observed individuals) and the results of the analysis are presented in Table 7. The most parsimonious model of the BMI inheritance was the simplest one among the three here analyzed traits, described by only 5 parameters. Namely, the most parsimonious MG model for BMI shows codominant MG effect on the trait value, no differences among residual variances in three genotypes and zero correlation between residuals in spouses and between parents and offspring. The hypotheses of the dominant and recessive MG control of the trait and the hypothesis that no MG takes place in the trait control, have been tested and statistically rejected. The accepted model explains smaller proportion of trait variance and is less predictive than the models obtained for body height and weight: H^2 = 0.394, D^2 = 0.406. Respective H^2_{BMI} and D^2_{BMI} values are 0.341 and 0.351. The prediction accuracy is measured by $R(x,\tilde{x})$ = 0.318, n = 874, for offspring; 0.235, n = 392, for parents; and 0.292, n = 1266, for the total sample.

TABLE 7
SEGREGATION ANALYSIS OF BMI (STANDARDIZED VALUES) IN PEDIGREE SAMPLE FROM MIDDLE DALMATIA, CROATIA

Parameter	General models			Most parsimonious models		
	General 1	Mendelian 2	Equal τ's 3	Arbitrary 4	Mendelian 5	Equal τ's 6
p	0.500	0.499	0.576	0.593	0.650±0.075	0.695
μ_1	–0.959	–1.029	–0.641	–0.668	–0.646±0.141	–0.410
μ_2	0.138	0.092	0.241	0.268#	0.280#	0.435#
μ_3	0.969	0.841	1.103	1.204	1.206±0.189	1.279
σ^2_1	0.452	0.431	0.633	0.587	0.600±0.067	0.713
σ^2_2	0.431	0.442	0.551	0.587!	0.600!	0.713!
σ^2_3	0.783	0.851	0.786	0.587!	0.600!	0.713!
ρ	0.181	0.167	0.060	[0.000]	[0.000]	[0.000]
β	0.003	–0.002	0.258	[0.000]	[0.000]	[0.000]
ε	0.193	0.177	0.159	0.191	0.187±0.051	0.246
τ_1	0.942	[1.000]	0.662	1.000+	[1.000]	0.783
τ_2	0.625	[0.500]	0.662!	0.630	[0.500]	0.783!
τ_3	0.000+	[0.000]	0.662!	0.000+	[0.000]	0.783!
LogLH	–1585.15	–1587.12	–1593.80	–1589.15	–1591.46	–1600.30
χ^2	–	3.94 ns (1; df=3)	17.30 ** (1; df=2)	8.00 ns (1; df=5)	4.62 ns (4; df=3)	22.30 ** (4; df=2)

ns p > 0.05; * p < 0.05; ** p < 0.01; (N) = number indicating the comparative column; [ ] parameter is fixed to shown value; ! parameter is constrained to be equal to the parameter above in the Table; # model is codominant; + parameter estimate achieved its limit.

On heterogeneity of the results

Because the analyzed sample represented a pool of pedigree data from 4 sub-populations, and because there were found some significant differences between the sub-populations in the studied traits (Table 1), the problem of the sample heterogeneity should be at least addressed if not solved in its entirety.

Table 8 summarizes the results of segregation analyses performed for body height, weight and BMI in total sample and in each of four sub-populations separately. It was expected that the sub-samples are less informative due to their reduced size. Thus, to make the comparison of different models more reasonably, we used only one variance parameter for 3 genotypes, in all cases when the more parsimonious equal-variance model was statistically accepted, as was the case for 9 analyses from 12. For weight in Hvar and Pelješac and for BMI in Brač three variances were used. Table 8 presents values of the transmission probability tests χ^2_3 and χ^2_2. It shows that the Mendelian transmission has been accepted in all 12 analyses. At the same time, equal-τ model has been rejected in all four subsamples only for body height. These results are quite understandable, bearing in mind that the power of the used tests was decreased substantially due to the reduced size of analyzed samples.

Table 9 provides estimations of the proportions of the traits variances (after adjustment for sex and age) attributable for the putative MG effect (H^2) and for all effects explicitly included in the MG model (D^2), for each sub-population and for

TABLE 8
χ^2-VALUES FROM THE TRANSMISSION PROBABILITY TESTS FOR BODY HEIGHT, WEIGHT AND BMI IN 4 SUB-SAMPLES AND IN TOTAL PEDIGREE SAMPLE (AGE & SEX ADJUSTED DATA)

Sample	Height			Weight			BMI		
	N	Mendelian	E-tau	N	Mendelian	E-tau	N	Mendelian	E-tau
Brač	473	0.03 ns	10.90*	457	7.27 ns	24.87**	453	5.52 ns	16.62**
Hvar	209	6.92 ns	12.75**	205	0.02 ns	20.21**	205	0.72 ns	0.86 ns
Korčula	272	5.13 ns	25.53**	198	3.71 ns	3.83 ns	199	3.41 ns	4.21 ns
Pelješac	300	5.20 ns	11.03*	294	1.51 ns	10.37*	291	2.01 ns	4.42 ns
Total sample	1254	3.36 ns	41.88**	1154	2.14 ns	22.62**	1148	3.94 ns	17.30**

ns = p > 0.05; * = p < 0.05; ** = p < 0.01

TABLE 9
PROPORTIONS OF BODY HEIGHT, WEIGHT AND BMI VARIANCES ATTRIBUTABLE TO MG EFFECT (H^2) AND TO ALL EFFECTS (D^2) INCLUDED IN THE NON-CONSTRAINED MENDELIAN MODEL IN 4 SUB-SAMPLES AND IN TOTAL PEDIGREE SAMPLE (AGE & SEX ADJUSTED DATA)

Sample	Height			Weight			BMI		
	N	H^2	D^2	N	H^2	D^2	N	H^2	D^2
Brač	473	0.443	0.532	457	0.649	0.696	453	0.548	0.593
Hvar	209	0.678	0.702	205	0.627	0.698	205	0.539	0.802
Korčula	272	0.430	0.626	198	0.378	0.449	199	0.592	0.828
Pelješac	300	0.578	0.623	294	0.595	0.614	291	0.505	0.516
Total sample	1254	0.505	0.579	1154	0.421	0.485	1148	0.451	0.460

the total sample. These proportions are found for the corresponding Mendelian models. As seen, these characteristics of fitting the MG model for the sub-samples are in most cases larger then they are for the total sample. This effect is well known: the model parameters can be better adjusted for each particular sub-sample data being less variable that the total sample. However, together with Table 8, these results seem to provide no evident difference between the sub-samples regarding the main problem considered here, the possibility to describe the inheritance of the studied traits in terms of the MG model.

Discussion

In the process of the most parsimonious model construction, all possible parameter constraints have been tested and, if they are not shown in the corresponding tables, it means that they were statistically rejected. In particular it is important to note that the multifactorial model describing the trait inheritance without any major gene effect was rejected for all three traits. Taking this into account, the presented results of segregation analysis clearly show that the inheritance of body height, weight and BMI, in Middle Dalmatian islander population, can be described in terms of a MG model. Along with the previously reported results[1], it makes six ethnically and geographically different populations where MG model of inheritance has been accepted for each of the three basic morphological features. It can be discussed as follows.

Shortly after Elston and Stewart (1971)[48] had proposed the modern technique of segregation analysis on pedigree samples, the first publication on the power of the transmission probability tests appeared (Go et al., 1978)[52], and these power studies have continued to appear up to the present time (see for example, Hodge, 1995)[53], in parallel with increasing complexity of the used MG models. It seems that the complex segregation analysis used here can be considered as a reliable and robust tool for a preliminary selection of traits controlled by a large-effect gene, which was well argued in the review of Jarvik (1998)[54].

We would like to note that the specific manner in which the »outlying« observations were defined here and excluded from the analyzed sample does not mean a forced acceptance of the MG model of the trait inheritance. Even for a one-dimensional sample, there is no widely accepted definition of the distribution tails containing the outlying observations. The limits of 3SD, 4SD etc., accepted in various publications are only the arbitrary choice of their authors justified mostly by tradition. These authors considered the »normal« observation as the one not substantially different from the trait mean. Considering a two-variable sample, it seems reasonable to define a »norm« not only for variation of each variable, but for their co-variation as well. It is quite evident that the pedigree sample cannot be specified as the one-dimensional because the sampled objects have their inner structure which itself is one of the important characteristics of the sampling procedure. For such a sample, it seems reasonable to find another definition of the »normal« observation. Here, we defined it as the observation compatible with the very idea of inheritance, though not necessary the mono or major gene one. The inheritance means larger trait similarity in members of the same pedigree, especially in first-degree relatives, than in individuals randomly chosen from the population. From this point of view, the definition of the outlying observations as those having difference in the trait values between the first-degree relatives exceeding 3SD seems quite justified, taking into

account that the standard deviation was found for the initial measurements of each trait. It should be noted, however, that the very fact of existence of the »outlying observations« could be interpreted also as evidence for certain heterogeneity of the studied populations. In this case, the reported results can be considered as the possibility to describe the studied trait inheritance in the MG model terms for the overwhelming majority of each population.

Thus, the acceptance of the MG model as a satisfactory description of inheritance for body height, weight and BMI, in six ethnically different populations seems worthy of being taken as a reliable result. At the same time, biological anthropology looks not for the satisfactory description of a human trait inheritance, but for the nature of this inheritance comparing genetic and environmental factors affecting the trait in various populations (Rogers et al., 1999)[55]. Concerning the obtained results, it seems unreasonable to assume that only one gene takes a part in the control of any of here investigated traits, and additionally, that the same gene plays the major role in all six populations. More probably, each of these traits is under control of several genes of different effect, and the gene with the largest effect can be responsible for the MG model acceptance, while the combined effect of other genes, together with environmental family effects, determine the non-MG effects estimated in the model. Since environmental conditions, as well as gene pools (and consecutively gene-gene and gene-environment interactions) are not uniformly shared in all human populations, it could be expected that different genes play the major role in the trait control in different populations. Thus, the acceptance of MG mode of inheritance of the same trait in different populations in reality may be the reflection of the action of different large-effect genes. The same is true for each of the sub-populations from which the pedigrees were collected in the present study. Uniting not overlapped pedigrees from 4 sub-samples cannot lead to recognizing 4 different MGs from a single common one.

Let us stress once more that this study, as well as our previous one, was intended only to tests whether inheritance of the three main human morphological characteristics can be satisfactorily described in terms of a major gene model. It appears that the answer is positive and that the obtained results are in agreement with previous studies dealing with inheritance of these traits mentioned in Introduction. However, we believe that on the present level of investigation, any comparison of particular parameter estimates and of magnitude of the MG-effects found in different studies is rather premature. We consider that any interpretation of the secondary results – obtained by putting constraints on the model parameters – could hardly be justified, taking into account a lot of specific sampling factors affecting these results.

Presented results, thus, justify further attempts in recognizing the large-effect genes supposedly taking part in the control of body height, weight and BMI. It seems that the only way of testing whether the above interpretation of the obtained results is realistic, and whether the same large-effect gene is causing the MG model acceptance in all populations – would be to perform a linkage analysis of the pedigree data.

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ANALIZA KOMPLEKSNE SEGREGACIJE VISINE TIJELA, MASE TIJELA TE BMI KORIŠTENJEM PODATAKA OBITELJI IZ SREDNJE DALMACIJE, HRVATSKA

S A Ž E T A K

Prema izvještaju Ginsburga i suradnika (1998)[1] koji su testirali podatke pet etnički i zemljopisno različitih populacija, način nasljeđivanja visine tijela, tjelesne mase te BMI moguće je opisati major genskim modelom (MG). U ovom radu, koristeći uzorak obitelji otočkih populacija Srednje Dalmacije, Hrvatska (1312 mjerenih ispitanika, članova 462 obitelji) prikazani su dokazi koji podupiru prethodne nalaze. Primjenom uobičajenih testova vjerojatnosti nasljeđivanja, prihvaćena je hipoteza prema kojoj se značajan dio varijacija svake od triju temeljnih morfoloških osobina može pripisati učinku jednog pretpostavljenog gena velikog učinka. Taj pretpostavljeni gen (major gen) odgovoran je za 39–50% varijacija ovih osobina (uz otklonjene učinke dobi i spola), odnosno za 34–48% ukupnih (za dob neporavnanih) varijacija visine, tjelesne mase i BMI.