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GENETIC DIFFERENTIATION OF UKRANIAN CHICKEN BREEDS USING VARIOUS TYPES OF MOLECULAR GENETIC MARKERS

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Abstract

Modern poultry breeding is aimed towards maximizing productive performance and genetic potential of chicken breeds and lines used for different purposes in order to obtain the greatest profit. Prevalence of foreign highly productive commercial chicken lines and crosses is determined by several factors, the most important of which are the high productivity of chicken lines, as well as the lack of support and ineffective implementation of programs targeted to genetic conservation of native breeds. Preferences given to highly productive chicken breeds in breeding and poultry farming also have negative effects which manifest in a reduced genetic diversity due to narrow specialization of selected breeds and lead to the reduction of national genetic resources. The study of genetically determined features of different chicken breeds is one of the priority tasks of the gene pool conservation problem. In this study, we used two types of molecular genetic markers, PCR-RFLP and Indel, to investigate the genetic differentiation of Ukrainian chicken breeds in comparative aspect based on polymorphism of different functional genes whose allelic variants are associated with productive traits. The Ukrainian chicken breeds for different primary use, i.e. Borkovskaya Barvistaya line A, Plymouth Rock White line G-2, Poltava clay line 14 and Rhode Island Red line 38, were compared. Genetic differentiation of the chicken populations was performed by analyzing frequencies of alleles in polymorphic loci of prolactin gene (*PRL*), growth hormone gene (*GH*), insulin-like growth factor I gene (*IGF-I*), gene family of transforming growth factors β (*TGF-β1*, *TGF-β2* and *TGF-β3*), pituitary transcription factor-1 gene (*PIT-1*) and Mx gene (*Mx*). For generalized estimation of breed diversity, the genetic distances were calculated based on the studied polymorphic loci for both PCR-RFLP and Indel markers. The most genetically distant breeds were Borkovskaya Barvistaya and Rhode Island Red (24.9 % of the differences). In general, the largest differences can be noted between the egg-lying and dual-purpose chicken breeds. In this, the allelic differences with the lines used for both eggs and meat were most pronounced (23-25 %). Differences between the breeds of dual use, i.e. primary for meat and eggs or for eggs and meat, were not expressed enough. Maximum differences were between populations of Poltava clay and Plymouth Rock White chicken (11.2 %), while minimum differences were between Rhode Island Red and White Plymouth Rock chicken (4.2 %). In turn, the genetic distance between the two egg-meat breeds studied was intermediate compared to the above-mentioned (7.1 % difference). The pattern of phylogenetic tree corresponds to the previously described regularities and reflects differentiation of the chicken lines by their primary use. As follows from the dendrogram, the chickens of egg-meat primary use form a separate cluster. At the same time, meat-egg and egg-lying chickens form separate branches, while the egg-lying breed shows the greatest genetic differences compared to the other lines.

Keywords: polymorphism, allele, population, chicken, genetic distances, egg chicken breeds, dual-purpose chicken breeds

Modern poultry breeding is aimed towards maximizing the potential use of chicken breeds and lines used for different purpose in order to obtain the greatest profit from sale of poultry. Worldwide spread of foreign highly productive commercial chicken crosses and lines depend on the several factors, the most important of which is high productivity values in poultry, as well as lack of

support and effective realization of the programs for conservation of genetic sources of the domestic breeds in general. Endless hurry for profit may often literally result in extermination of the breeds in creation of which decades of work were spent by the national geneticists and animal breeders. The key concept of modern poultry breeding industry is effectiveness that is expressed in the growing values of poultry yield [1]. However breeding of highly productive poultry may also have negative effects manifested in reduced genetic diversity because of narrow specialization of breeds and lines, leading to a decrease in national genetic resources [2, 3]. Lower interest in various genetic resources endangers their existence in general and may lead to loss of the unique genetic properties, which are alien to modern industrial poultry and which are characterizing local breeding groups in particular [4, 5].

At beginning of 1990s, the Poultry Breeding Institute at the Ukrainian Academy of Agrarian Sciences had tenths of agricultural poultry breeds and lines used for different purpose, including rare breeds — Yurlovo Crower, Italian partridge, bare-necked chicken breeds, mini chickens, etc. [6]. Today, the State Trial Poultry Breeding Station (successor of the above-named poultry breeding institute) has the limited number of the Ukrainian chicken breeds represented by only several lines. The most spread representatives of such "genetic core" are breeds of egg laying chickens — Borkovskaya Barvistaya (line A), meat-egg chickens — Plymouth Rock White (line G-2), dual-purpose chickens — Poltava clay (line 14), and Rhode Island Red (line 38 and line 02). Recently, the above-listed chicken breeds lack the importance for the industrial poultry breeding and are sold only for the needs of small farming units, which is to the most extent defined by good adaptive properties of the Ukrainian chicken upon keeping them at the courtyard. Lack of the expressed state support and interest of the large poultry producers in Ukraine endanger the existence of the genetic breeds in general, which, in case of their extermination, would result in permanent loss of the unique genetic material adapted to keeping conditions in relevant geographic zone. Use of genetic properties of various poultry breeds refers to priority genetic conservation issues [7]. Therefore, analysis of specific properties of the genetic structure of the Ukrainian chicken populations (along with genetic conservation in general) becomes the paramount task for the Ukrainian poultry breeding industry.

We have already studied genetic and population parameters of trial chicken lines. In this publication we for the first time put an emphasis on the genetic differentiation of selected Ukrainian populations in the comparable aspect based on data on polymorphism of various functional genes, allele variants of which are associated with emergence of the economically useful traits.

Purpose of the present study is molecular and genetic differentiation of the Ukrainian chicken breeds.

Technique. The studies of Ukrainian trial chicken populations including Borkovskaya Barvistaya (line A) egg-laying chickens, Plymouth Rock White (line G-2) meat-egg chickens, and Poltava clay (line 14) and Rhode Island Red (line 38) dual purpose chickens were carried out from 2011 to 2015.

Polymorphism of target genes by PCR-RFLP and Indel markers was studied. These were 57 bps insertion in intron 2 of *PIT-1* (gene of pituitary transcription factor-1); 24 bps insertion in promoter area and transition of cytosine to thiamin in position –2402 of *PRL* (prolactin gene); MspI polymorphism in intron 1 and intron 4, and SacI and AluI polymorphisms in intron 4 of *GH* (growth hormone gene); HinfI polymorphism in promoter area and PstI polymorphism in 5′UTR region of *IGF-I* (gene of insulin-like growth factor I); MboII polymorphism of *TGF-β1* (gene of transformation growth factor β1) exon

area; RsaI polymorphism of *TGF-β2* (gene of transformation growth factor β2) promoter area; BslI polymorphism in intron 4 of *TGF-β3* (gene of transformation growth factor β3); RsaI polymorphism in exon 13 of *Mx* (gene Mx).

The primers, protocols, and restriction enzymes were as described [8-15].

Amplification was done with the use of DreamTaq PCR Master Mix reagents (Thermo Scientific, USA) and a programmed thermal cycler TherCyc (DNA Technology, Russia) as per the protocol: denaturation for 5 min at 94 °C (1 cycle); denaturation for 1 min at 94 °C, annealing for 1 min at the temperature specific for each locus, elongation for 1 min at 72 °C (35 cycles); final elongation for 10 min at 72 °C (1 cycle). The final mixture volume was 20 μl, and concentration of primers was 0.2 μM. Genotyping was based on electrophoretic analysis.

Polymorphic allele frequency was determined by maximum likelihood formulas [16]. Based on the obtained data, the Nei genetic distances and Wright F-statistics were calculated by common methods with the use of Popgen32 software (https://sites.ualberta.ca/~fyeh/popgene_do-wnload.html). Divergence degree between the populations was determined based on F_{st}, with F_{st} of 0.00-0.05 for poor divergence, of 0.06-0.15 for medium divergence, of 0.16-0.25 for high divergence, and of > 0.25 for ultrahigh divergence [17]. Philogenetic tree was plotted using PHILIP 3.69 (http://evolution.gs.washington.edu/phylip/getmenew1.html) and MEGA 7 (https://www.megasoftware.net/download_form) softwares. Validity of allele frequency values and confidence limits of their diversity were determined by statistical error and t-test [16]. The differences were statistically significant at $p < 0.05$.

Results. Use of PCR method and restriction analysis enabled us to determine polymorphous variants of the selected genes in the Ukrainian chicken breeds. Structure of the primers, relevant restriction enzymes, as well as relative sizes of the amplification and restriction products are provided in table 1 below.

1. Nucleotide sequences of primers, relevant restriction enzymes, and relative sizes of the amplification/restriction products

Locus	Nucleotide sequences of primers (references)	Annealing	Restriction endonuclease	Amplification/restriction products, bps
PIT-1 (intron 2)	gtcaaggcaaatattctgtacc; tgcatgttaatttggctctg [8]	58 °C		I — 387; D — 330
PRL (promoter)	tttaatattgtgggtgaagagaca; atgccactgatcctcgaaaactc [9]	54 °C		I — 154; D — 130
PRL (C-2402T)	agaggcagcccaggcattttac; cctgggtctggtttggaaattg [9]	62 °C	AluI	C — 160/144/81/54; T — 304/81/54
GH (intron 1)	atccccaggcaaacatcctc; cctcgacatccagctcacat [10]	55 °C	MspI	A — 539/237; B — 392/237/147; C — 267/237/147/125
GH (intron 4)	ctaaaggacctggaagaaggg; aacttgtcgtaggtgggtctg [10]	61 °C	MspI	A — 1200; B — 600/600; C — 500/700
GH (intron 4)	ctaaaggacctggaagaaggg; aacttgtcgtaggtgggtctg [10]	61 °C	SacI	A —584/440/144; B — 1024/144
GH (intron 4)	ctgagggacgtggttatgggcac; gacctcaaggattgcagggct [11]	63 °C	AluI	C — 167/293; T — 108/185/167
IGF-1 (promoter)	cattgcgcaggctctatctg; tcaagagaagcccttca [12]	55 °C	HinfI	C —622/191; A — 378/244/191
IGF-1 (5′UTR)	gactatacagaaagaaccac; tatcactcaagtggctcaagt [13]	53 °C	PstI	C_1 — 621; C_2 — 257/364
TGF-β1 (exon)	ggggtcttcaagctgagcgt; ttggcaatgctctgcatgtc [14]	65 °C	MboII	B — 173/67; F — 240
TGF-β2 (promoter)	gccataggttcagtgcaag; tgacagaagctctcaagcc [14]	52 °C	RsaI	B — 100/184; L — 284
TGF-β3 (intron 4)	tcagggcaggtagagggtgt; gccactggcaggattctcac [14]	64 °C	BslI	B — 125/75/74/20; L — 145/75/74
Mx (exon 13)	ccttcagcctgtttttctcctttaggaa; cagaggaatctgattgctcaggcgtgta [15]	60 °C	RsaI	A — 100; G — 73/27

Table 2 shows allele frequencyies of the studied loci in the lines.

2. Allele frequencies of the studied loci in the populations of Ukrainian chicken breeds

Locus, restriction endonuclease	Breeds			
	Plymouth Rock White	Borkovskaya Barvistaya	Poltava clay	Rhode Island Red
PRL	0,135 (I)^{a};	0,710 (I)^{b};	0 (I)^{c};	0,060 (I)^{d};
24 Indel	0,865 (D)^{a}	0,290 (D)^{b}	1 (D)^{c}	0,940 (D)^{d}
PRL	0,155 (C)^{a};	0,710 (C)^{b};	0,372 (C)^{c};	0,140 (C)^{ad};
C-2402T	0,845 (T)^{a}	0,290 (T)^{b}	0,628 (T)^{c}	0,860 (T)^{ad}
GH	0,435 (A)^{a};	0,650 (A)^{b};	0,908 (A)^{c};	0,390 (A)^{ad};
intron 1	0,395 (B)^{a};	0,270 (B)^{b};	0,020 (B)^{c};	0,130 (B)^{d};
MspI	0,170 (C)^{a}	0,080 (C)^{b}	0,072 (C)^{bc}	0,480 (C)^{d}
GH	0,560 (A)^{a};	0,750 (A)^{b};	0,100 (A)^{c};	0,270 (A)^{d};
intron 4	0,160 (B)^{a};	0,080 (B)^{b};	0,070 (B)^{bc};	0,310 (B)^{d};
MspI	0,280 (C)^{a}	0,170 (C)^{b}	0,830 (C)^{c}	0,420 (C)^{d}
GH	0,030 (A)^{a};	0,550 (A)^{b};	0,036 (A)ac;	0,110 (A)^{d};
intron 4	0,970 (B)^{a}	0,450 (B)^{b}	0,964 (B)^{ac}	0,890 (B)^{d}
SacI				
GH	0,140 (C)^{a};	0,080 (C)^{ab};	0,040 (C)^{bc};	0,300 (C)^{d};
intron 4	0,860 (T)^{a}	0,920 (T)^{ab}	0,960 (T)^{bc}	0,700 (T)^{d}
AluI				
IGF-I	0,180 (C_1)^{a};	0,270 (C_1)^{b};	0,380 (C_1)^{c};	0,350 (C_1)^{bcd};
PstI	0,820 (C_2)^{a}	0,730 (C_2)^{b}	0,620 (C_2)^{c}	0,650 (C_2)^{bcd}
IGF-I	0,680 (A)^{a};	0,270 (A)^{b};	0,290 (A)^{bc};	0,420 (A)^{d};
HinfI	0,320 (C)^{a}	0,730 (C)^{b}	0,710 (C)^{bc}	0,580 (C)^{d}
TGF-β1	0,210 (B)^{a};	0,540 (B)^{b};	0,310 (B)^{c};	0,150 (B)^{ad};
	0,790 (F)^{a}	0,460 (F)^{b}	0,690 (F)^{c}	0,850 (F)^{ad}
TGF-β2	0,460 (B)^{a};	0,600 (B)^{b};	0,790 (B)^{c};	0,610 (B)^{bd};
	0,540 (L)^{a}	0,400 (L)^{b}	0,210 (L)^{c}	0,390 (L)^{bd}
TGF-β3	0,240 (B)^{a};	0,170 (B)^{ab};	0,520 (B)^{c};	0,330 (B)^{d};
	0,760 (L)^{a}	0,830 (L)^{ab}	0,480 (L)^{c}	0,670 (L)^{d}
Mx	0,210 (A)^{a};	0,375 (A)^{b};	0,140 (A)^{ac};	0,125 (A)^{cd};
	0,790 (G)^{a}	0,625 (G)^{b}	0,860 (G)^{ac}	0,875 (G)^{cd}
PIT-1	0,520 (I)^{a};	0,360 (I)^{b};	0,630 (I)^{c};	0,650 (I)^{cd};
	0,480 (D)^{a}	0,640 (D)^{b}	0,370 (D)^{c}	0,350 (D)^{cd}

Note. Sizes of amplification/restriction products for each studied locus (A, B, C, C_1, C_2, D, F, G, I, L, T) are provided in table 1; different letters in the upper index (a, b, c, d) signify the statistically significant differences ($p < 0.05$) within the locus limit.

Insertion in the promoter of prolactin gene indicates the expressed prevailing of allele I in the population of egg-laying chickens. The dual-purpose chickens (both egg and meat, and meat-egg) show prevailing of allele D, whereas in Poltava chickens this locus is monomorphic, i.e. the population entirely consists of individuals with DD genotype. Distribution of alleles for C-2402T mutation in *PRL* locus was somewhat similar. Thus, frequencies of C and T alleles in the population of egg-laying chickens were similar to those for I and D alleles. This interesting phenomenon is due to practically absolute prevalence of IC haplotype over IT haplotype, and DT haplotype over DC haplotype in the said population. Herewith, such trend was not observed in other populations. We assume that prevalence of IC haplotypes in egg-laying chickens directly reflects the effect of performed selection. Such assumption is confirmed by many authors who report on the relationship of I and C alleles with egg yield in chickens of various breeds that, in its turn, correlates with results of our studies [9, 18, 19]. Note, for this mutation, unlike the above-described, prolactin locus was polymorphic in the Poltava chicken population. Herewith, C allele frequency in line 14 was the highest for the dual-purpose chicken populations. In its turn, lines G-2 and 38 were practically identical by proportion of C and T allele frequencies.

Chicken lines for different primary use significantly differ in MspI polymorphism in intron 1 of growth hormone gene. Thus, Rhode Island Red line was characterized by prevalence of C allele (0.480), while its frequency was small in other populations. It should be noted that the smallest C allele frequency was in Poltava clay line (0.072), which just like Rhode Island Red line refers to dual-

purpose type. Possibly, the interbreed differences were more important than productivity types. At the same time, Poltava chickens were characterized by the highest A allele frequency (0.908) and the smallest B allele frequency (0.020) as compared to other studied lines. At that, only A allele was found homozygous in the said population. According to results of foreign authors, C allele is completely absent in the commercial lines of egg-laying chickens (Hy-Line), whereas its frequency in the native populations expressly varies [20]. Chicken lines of different primary use also significantly differ in allele frequencies on MspI polymorphism of intron 4 of growth hormone gene. Thus, no BB homozygotes were found in the Poltava clay and Borkovskaya Barvistaya populations. This results in low frequency of B allele. Herewith, Poltava clay line has the highest frequency of CC genotypes and, accordingly, the highest frequency of C allele. The highest frequency of allele A was in egg-laying chickens, and the smallest one was in dual-purpose chickens (line 14). Line 38 has the highest frequency of allele B. Egg-laying chickens expressly differed from other birds by SacI polymorphism of intron 4 of growth hormone gene due to prevalence of allele A (0.550). In other populations allele A was significantly less frequent, from 0.030 to 0.110, and found only in heterozygotes. Differences between lines for AluI polymorphism in intron 4 of growth hormone gene are sufficiently smoothed, except for the Rhode Island Red population with the highest allele C frequency. Allele T is expressly dominating in other populations.

Allele C_2 prevails for insulin-like growth factor I locus in all populations studied. This is mostly expressed in line G-2 of Plymouth Rock White (only one bird homozygous for allele C_1 was found). The closest allele frequencies were in chickens used for egg and meat. Intermediate position is characteristic of egg-laying chickens. Foreign researchers report that commercial meat chicken crosses have a significant prevalence of allele C_1 (for Cobb 500, C_1 frequency is 0.84) or sufficiently close ratio of allele frequencies (for Hubbard, C_1 frequency is 0.42) [21]. Other studies show high egg yield of C_2C_2 individuals compared to C_1C_1 ones in populations of the native Korean and Chinese breeds [22, 23]. The dual-purpose chicken lines have the highest differences from other studied populations by HinfI polymorphism of insulin-like growth factor I gene promoter area. This population was characterized by prevalence of AA homozygotes, which results in prevalence of relevant allele. The other populations did not practically differ from each other. These data correlate with foreign study results showing association of allele A in poultry with meat properties [12, 24]. In fact, dual-purpose chicken lines have higher meat properties (i.e. live weight, carcass weight, etc.) as compared to other breeds. Therefore, the observed distribution of allele frequencies in the studied populations is quite reasonable, but further studies are necessary to found out the links of *IGF-I* allele variants with productive properties of Plymouth Rock White line, given breed specificity of molecular markers.

The studied population of egg-laying chickens is leading on MboII polymorphism of *TGF-β1* exon area with the prevalence of allele B and maximum number of BB homozygotes. Allele F significantly prevails among chicken for different primary use with maximum frequency in the Rhode Island Red population which lacks BB homozygotes. It should be noted that during studies of the productive properties of chickens depending on allele variants *TGF-β1*, positive association of allele F with meat yield values was in Poltava clay breed that, in its turn, allows us to explain the observed allele frequencies [25]. As to RsaI polymorphism of *TGF-β2* promoter, we have not found specially expressed differences between the lines. In general, allele B prevails, except for Plymouth Rock White breed, with maximum frequency for Poltava chicken line which also has the

lowest heterozygosity. The proportion of *TGF-β3* alleles in populations of dual-purpose and egg-lying chickens was practically the same (no valid differences were found) with significant prevalence of allele L. Line 38 was denoted by practically double prevalence of allele L regarding B. Differences in allele frequencies in line 38 compared to other populations were valid. Population of Poltava clay chickens in which frequencies of B and L alleles have practically coincided (0.520 vs. 0.480) validly differed from other studied lines.

Similar RsaI polymorphism for gene *Mx* was denoted in all populations. Allele G prevailed over allele A that was mainly expressed in egg-and-meat chicken and the least expressed in egg-laying chickens. Moreover, it is egg-laying chicken line where we have found the greatest number of AA homozygotes. By *PIT-1* allele ratio, egg-lying and egg-and-meat chickens significantly differed while dual-purpose breeds are the intermediates (see Table 2).

For generalized assessment of the genetic differentiation in the populations of chickens for different primary use, we have estimated genetic distances based on the studied polymorphic loci (both PCR-RFLP and Indel markers were analyzed). The values and the opposite genetic likelihood indices are provided in the Table 3.

3. Genetic distances and genetic likelihood between the the populations of the Ukrainian chicken breeds

Breed	Plymouth Rock White	Borkovskaya Barvistaya	Poltava clay	Rhode Island Red
Plymouth Rock White		0.1951	0.1124	0.0424
Borkovskaya Barvistaya	0.8228		0.2319	0.2488
Poltava clay	0.8937	0.7930		0.0712
Rhode Island Red	0.9585	0.7797	0.9313	

N o t e. Genetic distance values are above the diagonal, genetic likelihood values are under the diagonal.

The most genetically apart breeds were Borkovskaya Barvistaya and Rhode Island Red (24.9 % of differences). In general, the least likelihood was found in egg-laying and dual-purpose breeds, whereas the least likelihood was noted in egg-meat chickens (23-25 % of differences in allele variants of loci) (see Table 3). Dual-purpose and egg-and-meat chickens differ slightly, Poltava clay and Plymouth Rock White lines are the most differentiated (11.2 %), Rhode Island Red and Plymouth Rock White are the least differentiated (4.2 %). In turn, intermediate genetic distances were between the two breeds of egg-and-meat types (7.1 % differences).

Having analyzed the genetic differentiation in chicken populations, we could note specific distribution of allele frequencies of the studied polymorphic loci depending on the productivity type. In this, the extent of differences by different markers was not the same. In this regard, it is reasonable to assess the degree of dissimilarities between the populations by separate loci. In addition to estimation of the genetic distances by Nei, coefficient F_{st} may serve as a good instrument as it directly reflects subdivision of the populations and may be calculated for individual loci. F_{st} values for each locus within all studied populations may be used to estimate dissimilarities (genetic subdivision) in the chickens as per the selected genes.

Both studied mutations in prolactin locus clearly display the dissimilarities between the egg-laying and dual-purpose chickens as it is evidenced by F_{st} values from 0.34 to 0.55 for 24 Indel and from 0.11 to 0.33 for C-2402T, respectively. Maximum dissimilarities for the insertion in the prolactin locus were in line 14 due to monomorphism of this locus in the studied population. F_{st} deviations for dual-purpose lines were insignificant. Wright's genetic subdivision values (F_{st}) correlate with Nei's genetic distance (Dn) calculated separately for prolactin locus. Dn values for populations of egg-laying and dual-purpose chickens are from 0.66 to 0.97 for 24 Indel and from 0.22 to 0.65 for C-2402T.

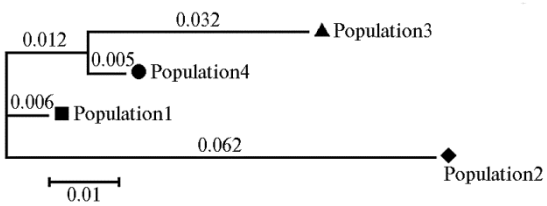
The allele frequencies of growth hormone locus are directly determined by mutation type. By intron 1 polymorphism (MspI-polymorphism), the Poltava clay population stands apart and clearly differ from the meet-and-egg lines (Fst = 0.19) and the egg-meet lines (Fst = 0.23) but not from the egg-laying lines (0.09). Dissimilarities between other lines were insignificant. Only the Rhode Island Red chickens are somewhat dissimilar from other studied populations by AluI polymorphism in intron 4, with F_{st} from 0.04 to 0.12 vs. F_{st} of 0.03-0.08. By SacI polymorphism in *GH* intron 4 compared to other mutations in this locus, the egg-laying and combines lines are the most dissimilar, with F_{st} of 0.23 to 0.33. Combined breeds have practically minimum F_{st} values of 0.02-0.08. The highest similarity by MspI polymorphism in *GH* intron 4 was between Plymouth Rock White and Borkovskaya Barvistaya breeds (F_{st} = 0.03), as well as Rhode Island Red breed (F_{st} = 0.05), while the last two breeds, in turn, differed from each other (F_{st} = 0.14). The Poltava clay and egg-laying chickens (F_{st} = 0.39), as well as Plymouth Rock White chickens (F_{st} = 0.23) are the most diverse. These trends completely correspond to the Nei's genetic distance.

Study of transforming growth factor β gene family polymorphism revealed that by MboII polymorphism of *TGF-β1* exon the egg-laying chicken population is the least diverse from line 14 (F_{st} = 0.11) and the most diverse from line 38 (F_{st} = 0.24) as compared to meet-egg chickens (F_{st} = 0.18). The populations for combined use do not differ. No expressed dissimilarities between the lines were found for RsaI polymorphism of *TGF-β2* promoter fragment. A comparison of Plymouth Rock White and Poltava clay populations results in the highest F_{st} value. F_{st} for locus *TGF-β3* in all populations also did not reflect significant dissimilarities.

We did not found any significant F_{st} distance between all populations for PstI polymorphism of insulin-like growth factor I locus. The meat-egg chicken line shows the highest HinfI polymorphism of insulin-like growth factor I gene promoter, except for comparisons with Rhode Island Red chickens. For all other lines dissimilarities are insignificant.

Also, we have not found any significant differences of F_{st} for *PIT-1* locus. RsaI polymorphism of *Mx* gene is similar for all populations, with F_{st} from 0.01 to 0.08.

In general, allele frequencies in chicken populations may depend on both selection purpose (towards egg or meat yield, etc.), and characteristic features of breeds. Moreover, in phenotypic-based selection of individuals for nest formation the breeder often uses several characteristics of which each is due to effects of several genes (alleles). Quantitative traits are determined by the aggregate activity of the significant number of genes and, thus, the selected individuals with desired performance parameters may also have nonproductive alleles especially manifested in heterozygotes. All these factors together result in the distribution we observed.



Dendrogram of interpopulation relationships by Nei's genetic distance (Neighbor Joining method): Population 1 — Plymouth Rock White, line G-2; Population 2 — Borkovskaya Barvistaya, line A; Population 3 — Poltava clay, line 14; Population 4 — Rhode Island Red, line 38.

Dendrogram of genetic diversity of the chicken populations (Fig.) was plotted by Neighbor Joining method with the use of genetic distances for all studied loci. The obtained phylogenetic tree generally corresponds to earlier described regularities and reflects the dissimilarities between the chicken lines de-

pending on the chicken types. Populations of egg-meat chickens are clustered together. At the same time, meet-and-egg and egg-laying chickens form separate branches, provided that the egg-laying breed demonstrates the highest genetic dissimilarities as compared to other lines.

Our results testify that, traditional phenotype-based selection in fact does not allow for the desired genotypes and elimination of individuals possessing nonproductive alleles of the wide range of loci. Traditional selection practice shows that Ukrainian breeds are inferior to the imported chicken lines, mainly because of limited use of marker associated selection (MAS) in poultry breeding. MAS is a quit routine tool enabling an increase in poultry productivity to the level of foreign lines with a number of monomorphic candidate genes, as it is shown by foreign researches.

Thus, our data validly prove that genetic differentiation of the chicken populations by a set of polymorphic loci mainly depends on the poultry primary use. However, the effect of this factor may vary. Genetic variability revealed in each of the studied breeds allows targeted selection with the use of molecular and genetic methods, including individual QTL genotyping, to produce the lines with a certain set of desired genotypes and their combinations. It will enable breeders to maximum use of the productive potential of the Ukrainian chicken breeds.

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