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## A Computer-Assisted Population Frequency Study of 14 Polymorphic Blood Grouping Systems in North Carolina

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**ABSTRACT:** Results of a population frequency study conducted on 14 polymorphic blood grouping systems in the North Carolina white and black populations are reported. A microcomputer program has been developed to facilitate the handling and storage of the large volume of data generated during the two years it took to complete this study. This computer program was also designed to calculate the combined phenotypic frequency for a given set of blood types.

**KEYWORDS:** forensic science, genetic typing, computers

The adversarial nature of courtroom testimony has undergone a significant change in the last decade, from a system relying primarily on eyewitness testimony and confession to an increasing reliance on circumstantial evidence and expert scientific testimony. This change has been brought about by the rapidly expanding capabilities of forensic scientists and by an increased sophistication of the courts, which have demanded scientists' participation in the examination of physical evidence. Forensic serologists have aided the courts by identifying individuals involved in crime through the identification and typing of body fluids spilled during the perpetration of the crime. When blood types from a questioned stain match the blood types of a known individual, the blood from the questioned stain has been shown to be consistent with having originated from that individual. Mathematical calculations can be used to determine the probability that another individual has the same combination of blood groups, which may be useful to the jury in considering how strongly to consider the evidence.

Several appeals of conviction, the latest being the *State of North Carolina v. L. D. Kirby [1]*, have been registered with the Appellate and Supreme Courts of North Carolina that challenge the use of population frequency data by North Carolina State Bureau of Investigation (NCSBI) serologists in court. In addition, numerous objections have been raised in North Carolina courts when individuals from our laboratory testified to population frequency data neither generated by our laboratory nor specific for the North Carolina population. To date, these objections have been overruled and all appeals have been denied.

In an effort to eliminate these objections and appeals, the NCSBI laboratory undertook a population survey for the state of North Carolina beginning in January 1981. In order to facilitate the handling of the large volume of data anticipated, a microcomputer program was designed to perform two basic tasks. The first task was to automatically sort and store

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blood typing data entered by blood grouping system, phenotype, and race. This allowed us to maintain a running total of the observed frequency and observed number of each phenotype in each blood grouping system for each race. The second task was to calculate the combined phenotypic frequency for a given set of blood types. This information was useful in writing laboratory reports and in preparing for court testimony on population frequency data.

Microcomputers have been used before this study for the calculation of population frequency data. Tanton [2] described a program to calculate the combined phenotypic frequency for a set of blood types for the three different races found in his jurisdiction. Wist [3] described a computer program to calculate the combined phenotypic frequency for a set of blood types and also to store and to retrieve data by case file number for comparative purposes. The portion of Wist's program dealing with the calculation of population frequency data was the forerunner of part of the program designed for this study.

## **Materials and Methods**

### *Computer Equipment Used*

The microcomputer used in this study was an Apple II Plus computer, which has 48K of random access memory. Peripheral equipment included a cathode ray tube (CRT) display, a disk drive, and an Apple Silent Type printer. Data were stored on a 16-sector, 5¼-in. floppy magnetic disk.

### *The NCSBI Serology Program*

The NCSBI Serology Program, written in Applesoft, is initiated by a password program designed to limit access to authorized personnel, thereby avoiding erroneous input of data. The remainder of the program is divided into the main program and its accompanying text file.

The main program allows one to select from six program options that perform two basic tasks. The first task is to calculate the combined phenotypic frequency for a set of blood types. In Option 1 (enter data and calculate statistics), one enters the observed phenotypes for each of the blood grouping systems tested and the racial origin of a given blood sample. After one verifies that all the entries are correct, the computer calculates the percent and number fraction of the population that has the same combination of blood types.

When this program was designed, our laboratory had to use population frequency data that were gleaned from a combination of sources across the country [4-8], since the North Carolina population frequency data were yet to be obtained. These population frequency data were used to perform the calculations in Option 1 of this program and were stored in the text file accompanying the main program. The population frequency data can be updated or changed at any time by using Program Option 3 (update U.S. population statistics). Now that a sufficient data base has been established for the North Carolina population, we have substituted our population frequency data (reported in this manuscript) in place of the "old" U.S. population frequency data. Similarly, any laboratory with access to this program could easily substitute their own population frequency data.

The second major task of this program is to file and store all blood typing information entered for North Carolina residents by blood grouping system, phenotype, and race. Each time an entry is made in Option 1, that entry is automatically added to the North Carolina population data base generated from previous entries, thereby keeping a running total of North Carolina population frequency information. Program Option 2 (retrieve North Carolina statistics) allows one to retrieve the latest cumulative North Carolina population frequency data generated by entries in Option 1. When Option 2 is used, the CRT displays the

observed number and frequency for each phenotype in each blood grouping system and for each race. A hard copy printout is also available for the latest North Carolina population frequency data. The North Carolina population frequency data retrieved by Option 2 can be edited by using Option 6 (change North Carolina statistics).

Since a population frequency survey should be composed of random, nonrelated individuals to remain unbiased, we devised procedures to ensure that the same individual was entered only once and that if several members of the same family were tested, only one individual's phenotypes were entered.

For those unfamiliar with microcomputer operations, we should explain that the programs and the data stored in them are supplied to the computer from a magnetic disk. Since the computer erases all memory of the program and its data after it is turned off, it is necessary to transmit (update) any new information in the computer to safe storage on the disk. Program Option 4 (update a second disk) and Program Option 5 (exit the program) both update the text file that contains the North Carolina population frequency data and the population statistics used in the calculations in Option 1. Option 4 performs this updating function on two disks, whereas Option 5 updates only one disk and then immediately exits the program.

### *Sources of Blood*

Blood samples used in this study were obtained from three sources. The first source was blood from victims and suspects of crime submitted to the NCSBI laboratory for typing tests. These samples were drawn via venipuncture into vacutainer tubes; in the case of homicide victims the blood was placed into clear plastic vials. Neither the vacutainer tubes nor the plastic vials contained preservatives or anticoagulants. Blood samples were also drawn from NCSBI employees by finger puncture onto clean cotton sheeting to form a stain, which was allowed to air dry. The final source of blood was the Virology and Serology Branch of the North Carolina Public Health Laboratory. These samples were obtained via venipuncture into vacutainer tubes that contained no preservatives or anticoagulants.

### *Methods of Blood Group Typing*

The ABO types were determined by the direct and indirect tube typing tests of the liquid blood. Lewis typing was by the direct tube method or the ficin-capillary tube method of Mudd [9], using Ortho Lewis a and Lewis b antisera prepared from goat serum. All protein and enzyme typing tests were performed on bloodstains that were prepared by pipetting whole blood onto clean cotton sheeting and allowing the bloodstains to air dry. Table 1 shows the gel type and electrophoretic method reference for each of the protein and enzyme systems examined.

### **Results of the North Carolina Population Survey**

North Carolina population frequency data for 13 blood grouping systems is shown in Tables 2 to 6. These results reflect similar values to previously published data from several different U.S. communities [4-8] as shown in Table 7. The hemoglobin, carbonic anhydrase, and peptidase A blood grouping systems were found to be monomorphic in the North Carolina white population and the adenosine deaminase system was found to be monomorphic in the North Carolina black population. In all other blood grouping systems, the North Carolina population frequency data fit Hardy-Weinberg equilibrium at a 5% level of significance as determined by chi square testing. The Yates correction factor was used to calculate chi square values for polymorphic systems having only one degree of freedom, since the accuracy of the determination is increased in cases where the values from either class are low.

TABLE 1—*Electrophoretic procedures used.*

Blood Grouping System	Gel Type	Reference
Erythrocyte acid phosphatase (EAP)	2 mm horizontal, 10% starch	[10]
Phosphoglucomutase (PGM)	1 mm horizontal, 10% starch	[11]
Haptoglobin (Hp)	4-21% vertical gradient polyacrylamide	[4]
Esterase D (EsD)	1 mm horizontal, 1% agarose-2% starch	[12]
Glyoxalase (GLO)	1 mm horizontal, 10% starch	[10]
Adenylate kinase (AK)	1 mm horizontal, 10% starch	[12]
Adenosine deaminase (ADA)	1 mm horizontal, 10% starch	[12]
Hemoglobin (Hb)	Titan III-H cellulose acetate	[13]
Carbonic anhydrase (CA)	1 mm horizontal, 1% agarose Type 2	[14]
Peptidase A (PepA)	1 mm horizontal, 1% agarose Type 2	[14]
6 Phosphogluconate dehydrogenase (6PGD)	1 mm horizontal, 10% starch	[12, 15]
Phosphoglucomutase subtyping (PGM sub)	0.5 mm polyacrylamide electrofocusing, pH 5.0-6.5	[16]

TABLE 2—*ABO and erythrocyte acid phosphatase (EAP): North Carolina population frequency data.*

Group	Type	Observed Number	Observed Incidence	Expected Incidence	Gene Frequency	$\chi^2$	
ABO	Whites	A	327	0.423	0.419	$ABO^A = 0.258$	0.075
		B	70	0.091	0.085	$ABO^B = 0.060$	
		O	356	0.460	0.465	$ABO^O = 0.682$	
		AB	20	0.026	0.031	...	
		<i>N</i>	773				
ABO	Blacks	A	155	0.241	0.249	$ABO^A = 0.159$	0.195
		B	132	0.206	0.213	$ABO^B = 0.138$	
		O	322	0.502	0.494	$ABO^O = 0.703$	
		AB	33	0.051	0.044	...	
		<i>N</i>	642				
EAP	Whites	A	49	0.117	0.098	$EAP^A = 0.313$	5.980
		B	169	0.404	0.385	$EAP^B = 0.620$	
		BA	144	0.345	0.388	$EAP^C = 0.067$	
		CB	36	0.086	0.083	...	
		CA	20	0.048	0.042	...	
		C	0	0.000	0.004	...	
		<i>N</i>	418				
	Blacks	A	16	0.044	0.039	$EAP^A = 0.197$	2.821
		B	220	0.601	0.591	$EAP^B = 0.769$	
		BA	106	0.290	0.303	$EAP^C = 0.011$	
		CB	4	0.011	0.017	$EAP^R = 0.023$	
		CA	2	0.005	0.004	...	
		C	0	0.000	0.000	...	
RA	4	0.011	0.009	...			
RB	13	0.035	0.035	...			
RC	0	0.000	0.001	...			
R	1	0.003	0.001	...			
<i>N</i>	366						

TABLE 3—Phosphoglucomutase (PGM) and phosphoglucomutase subtype (PGM sub): North Carolina population frequency data.

Group	Type	Observed Number	Observed Incidence	Expected Incidence	Gene Frequency	$\chi^2$
<b>PGM</b>						
Whites	1	496	0.596	0.598	$PGM^1 = 0.774$	0.020
	2-1	294	0.353	0.349	$PGM^2 = 0.226$	
	2	41	0.049	0.051	...	
	Other	2	0.002	0.002	...	
		<i>N</i> 833				
Blacks	1	504	0.649	0.648	$PGM^1 = 0.805$	0.065
	2-1	242	0.311	0.313	$PGM^2 = 0.195$	
	2	30	0.039	0.038	...	
	Other	1	0.001	0.001	...	
		<i>N</i> 777				
<b>PGM sub</b>						
Whites	1+1+	127	0.374	0.382	$PGM^{1+} = 0.618$	4.186
	1+1-	72	0.212	0.190	$PGM^{1-} = 0.154$	
	1-1-	6	0.018	0.024	$PGM^{2+} = 0.161$	
	2+2+	11	0.032	0.026	$PGM^{2-} = 0.067$	
	2+2-	9	0.026	0.022	...	
	2-2-	1	0.003	0.005	...	
	2+1+	67	0.197	0.199	...	
	2+1-	12	0.035	0.049	...	
	2-1+	27	0.079	0.083	...	
	2-1-	8	0.024	0.021	...	
			<i>N</i> 340			
Blacks	1+1+	151	0.429	0.419	$PGM^{1+} = 0.648$	4.969
	1+1-	61	0.173	0.185	$PGM^{1-} = 0.142$	
	1-1-	9	0.026	0.020	$PGM^{2+} = 0.163$	
	2+2+	12	0.034	0.027	$PGM^{2-} = 0.047$	
	2+2-	4	0.011	0.015	...	
	2-2-	1	0.003	0.002	...	
	2+1+	68	0.193	0.211	...	
	2+1-	19	0.054	0.047	...	
	2-1+	25	0.071	0.061	...	
	2-1-	2	0.006	0.013	...	
			<i>N</i> 352			

When two or more degrees of freedom occurred within a polymorphic blood grouping system, the Yates correction factor was not used [17].

Several interesting phenomena noted in the data in Tables 2 to 6 should be pointed out. The gene frequency of erythrocyte acid phosphatase (EAP)  $EAP^C$  in the North Carolina black population was extremely low ( $EAP^C = 0.011$ ). This phenomenon has been noted previously by other workers [4-8], who have assigned a gene frequency to  $EAP^C$  ranging from 0.000 to 0.0109 in the black populations they studied. The North Carolina white population, by contrast, gave an elevated  $EAP^C$  gene frequency value ( $EAP^C = 0.067$ ) over values in white populations studied by previous workers [4-8], which ranged from 0.024 to 0.061. The most surprising observation noted in the EAP results was how often the allele  $EAP^R$  appeared in the North Carolina black population ( $EAP^R = 0.023$ ). Stolorow et al [6] assigned the  $EAP^R$  allele a gene frequency of 0.0129 in their survey of blacks from Detroit, MI.

In this population survey, haptoglobin 0 (Hp 0) was not included since haptoglobin levels can be affected by disease, especially acute hemolysis [18]. Since we had no idea of the health

TABLE 4—Haptoglobin (*Hp*), esterase D (*EsD*), and glyoxalase (*GLO*): North Carolina population frequency data.

Group	Type	Observed Number	Observed Incidence	Expected Incidence	Gene Frequency	$\chi^2$
<b>Hp</b>						
Whites	1	64	0.162	0.171	$Hp^1 = 0.414$	0.467
	2-1	199	0.504	0.485	$Hp^2 = 0.586$	
	2-1M	0	0.000	0.000	...	
	2	132	0.334	0.344	...	
		<i>N</i> 395				
Blacks	1	91	0.287	0.287	$Hp^1 = 0.536$	0.003
	2-1	142	0.448	0.498	$Hp^2 = 0.464$	
	2-1M	16	0.050		...	
	2	68	0.215	0.215	...	
		<i>N</i> 317				
<b>EsD</b>						
Whites	1	312	0.804	0.806	$EsD^1 = 0.898$	0.058
	2-1	73	0.188	0.184	$EsD^2 = 0.102$	
	2	3	0.008	0.010	...	
		<i>N</i> 388				
Blacks	1	300	0.874	0.867	$EsD^1 = 0.931$	2.361
	2-1	39	0.114	0.128	$EsD^2 = 0.069$	
	2	4	0.012	0.005	...	
		<i>N</i> 343				
<b>GLO</b>						
Whites	1	53	0.171	0.185	$GLO^1 = 0.430$	0.609
	2-1	160	0.518	0.490	$GLO^2 = 0.570$	
	2	96	0.311	0.325	...	
		<i>N</i> 309				
Blacks	1	31	0.095	0.088	$GLO^1 = 0.297$	0.254
	2-1	132	0.404	0.418	$GLO^2 = 0.703$	
	2	164	0.501	0.494	...	
		<i>N</i> 327				

of our donors (indeed, some were of extremely poor or even terminal health), we typed Hp 0 individuals as giving inconclusive results and they did not enter into the calculation of the population frequency data shown in Table 4. Since the genetic control of the phenotype Hp 2-1M is unclear and may be explained by the existence of a separate allele  $Hp^{2M}$  or by a pair of controller-regulator genes acting on  $Hp^1$  and  $Hp^2$  [19], the Hp 2-1M phenotype was incorporated with the phenotype Hp 2-1 during the calculation of the gene frequencies of  $Hp^1$  and  $Hp^2$ .

Rare blood types have been entered as "other" in Tables 3, 5, and 6. The phosphoglucomutase ( $PGM_1$ ) "other" variants detected in the North Carolina white population were a  $PGM_1$  a6-1 found in an NCSBI employee (see Dykes et al [20] for a description of this variant) and a  $PGM_1$  3-1 found in a case sample [11]. The  $PGM_1$  "other" variant noted in the North Carolina black population was  $PGM_1$  6-1. In addition, three North Carolina black individuals were typed as being the "Atkinson" phenotype ( $PGM_2$  2-1). These samples were not entered as  $PGM_1$  "other" variants, since the variant allele occurs at a different PGM locus. Since three of the 777 North Carolina blacks tested have been typed as  $PGM_2$  2-1, the gene frequency of  $PGM_2^2$  in the North Carolina black population is 0.002 and the observed incidence of  $PGM_2$  2-1 is 0.004. The peptidase A (PepA) "other" variant noted in the North Carolina black population (Table 6) was a PepA 8-2. The six phosphogluconate dehydrogenase (6PGD) "other" variant shown in the North Carolina white population (Table 5) was a 6PGD AL (Lowell) variant. This new phenotype has been described in greater detail in a

TABLE 5—Adenylate kinase (AK), adenosine deaminase (ADA), and 6-phosphogluconate dehydrogenase (6PGD): North Carolina population frequency data.

Group	Type	Observed Number	Observed Incidence	Expected Incidence	Gene Frequency	$\chi^2$
<b>AK</b>						
Whites	1	414	0.937	0.934	$AK^1 = 0.9665$	2.170
	2-1	26	0.059	0.065	$AK^2 = 0.0335$	
	2	2	0.004	0.001	...	
		<i>N</i> 442				
Blacks	1	391	0.978	0.978	$AK^1 = 0.989$	0.012
	2-1	9	0.022	0.022	$AK^2 = 0.011$	
	2	0	0.000	0.000	...	
		<i>N</i> 400				
<b>ADA</b>						
Whites	1	389	0.892	0.895	$ADA^1 = 0.946$	0.583
	2-1	47	0.108	0.102	$ADA^2 = 0.054$	
	2	0	0.000	0.003	...	
		<i>N</i> 436				
Blacks	1	395	0.990	0.990	$ADA^1 = 0.995$	n.a.
	2-1	4	0.010	0.010	$ADA^2 = 0.005$	
	2	0	0.000	0.000	...	
		<i>N</i> 399				
<b>6PGD</b>						
Whites	A	305	0.962	0.959	$6PGD^A = 0.9795$	1.883
	AC	10	0.032	0.037	$6PGD^C = 0.0190$	
	C	1	0.003	0.001	...	
	other	1	0.003	0.003	...	
		<i>N</i> 317				
Blacks	A	281	0.909	0.919	$6PGD^A = 0.9545$	0.650
	AC	27	0.088	0.084	$6PGD^C = 0.0440$	
	C	0	0.000	0.002	...	
	other	1	0.003	0.003	...	
		<i>N</i> 309				

recent publication by Nelson [15]. The 6PGD "other" variant noted in the North Carolina black population was believed to be a 6PGD AR (Richmond) variant.

Table 8 compares the observed frequencies for the Lewis system in the North Carolina population with values reported by Molthan [21] from the Delaware Valley and with values reported by Miller et al [22] from New York. Although the values observed in this study are very similar to those shown by Molthan [21], both show elevated levels of Lewis a-b- individuals over the study of Miller et al [22]. These high levels of Lewis a-b- individuals may reflect the fact that the Lewis antigens are unstable on storage and undergo seasonal and individual variations [23], or that Lewis typing in this study and that of Molthan [21] was carried out by using antisera produced in goats, while Miller et al [22] used antisera derived from human sources.

In any case, the primary reason that Lewis typing data were collected was to obtain frequencies of secretors and nonsecretors in the North Carolina population. Lewis a+b- individuals are nonsecretors, Lewis a-b+ individuals are secretors, and Lewis a-b- individuals can be either secretors or nonsecretors. Since the Lewis and secretors genes (*Le*, *le*, *Se*, and *se*) each segregate independently, the secretor-to-nonsecretor ratio in individuals of the phenotype Lewis a-b- should reflect the secretor to nonsecretor ratio found in the rest of the population [23]. Using the information derived from the North Carolina Lewis data, 76.8% of the North Carolina white population were secretors and 23.2% were nonsecretors, whereas 71.4% of the North Carolina black population were secretors and 28.6% were non-

TABLE 6—Carbonic anhydrase (CA), peptidase A (PepA), and hemoglobin (Hb): North Carolina population frequency data.

Group	Type	Observed Number	Observed Incidence	Expected Incidence	Gene Frequency	$\chi^2$
<b>CA</b>						
Whites	1	334	0.997	0.997	$CA^1 = 0.9985$	n.a.
	2-1	1	0.003	0.003	$CA^2 = 0.0015$	
	2	0	0.000	0.000	...	
		<i>N</i> 335				
Blacks	1	313	0.792	0.789	$CA^1 = 0.8885$	0.070
	2-1	76	0.193	0.198	$CA^2 = 0.1115$	
	2	6	0.015	0.013	...	
		<i>N</i> 395				
<b>PepA</b>						
Whites	1	314	0.997	0.997	$PepA^1 = 0.9985$	n.a.
	2-1	1	0.003	0.003	$PepA^2 = 0.0015$	
	2	0	0.000	0.000	...	
		<i>N</i> 315				
Blacks	1	318	0.859	0.851	$PepA^1 = 0.9225$	3.047
	2-1	47	0.127	0.140	$PepA^2 = 0.0760$	
	2	4	0.011	0.006	...	
	other	1	0.003	0.003	...	
		<i>N</i> 370				
<b>Hb</b>						
Whites	A	365	1.000	1.000	$Hb^A = 1$	n.a.
		<i>N</i> 365				
Blacks	A	333	0.915	0.914	$Hb^A = 0.9560$	1.339
	AS	24	0.066	0.066	...	
	S	0	0.000	0.001	$Hb^S = 0.0345$	
	AC	6	0.016	0.018	$Hb^C = 0.0095$	
	SC	1	0.003	0.001	...	
	C	0	0.000	0.000	...	
		<i>N</i> 364				

secretors. These values are in good agreement with results reported by Zmijewski and Fletcher [23].

**Summary**

A computer program has been designed that quickly and accurately calculates the combined phenotypic frequency for a given set of blood types and that also files and stores the blood typing data entered with the express purpose of conducting a population frequency survey.

The traditional approach of performing population frequency surveys is to obtain several hundred blood samples at one time, analyze them, and then compile the data. The quantity of work and concentration of effort inherent in this approach usually prevents generally understaffed and backlogged forensic science laboratories from being able to conduct population surveys of their jurisdiction. Use of a computer program that can store and compile data over protracted periods of time will enable many laboratories to conduct population frequency surveys using data obtained from routine analysis of casework. A program like the one described in this study will enable laboratories to store and file population frequency data while entering their blood typing results to obtain combined phenotypic frequency information for inclusion in their laboratory reports.

Having North Carolina population frequency data for the blood group systems we cur-



TABLE 7—Comparison of gene frequencies from the North Carolina population with several other United States communities.<sup>a</sup>

Reference	Whites			Blacks					
	ABO								
	<i>ABO</i> <sup>A</sup>	<i>ABO</i> <sup>B</sup>	<i>ABO</i> <sup>O</sup>	<i>N</i>	<i>ABO</i> <sup>A</sup>	<i>ABO</i> <sup>B</sup>	<i>ABO</i> <sup>O</sup>	<i>N</i>	
NCSBI	0.258	0.060	0.682	773	0.159	0.138	0.703	642	
[7]	0.224	0.083	0.692	6004	0.168	0.142	0.690	1025	
[8]	0.262	0.074	0.664	914	0.166	0.127	0.707	713	
EAP									
	<i>EAP</i> <sup>A</sup>	<i>EAP</i> <sup>B</sup>	<i>EAP</i> <sup>C</sup>	<i>N</i>	<i>EAP</i> <sup>A</sup>	<i>EAP</i> <sup>B</sup>	<i>EAP</i> <sup>C</sup>	<i>EAP</i> <sup>R</sup>	<i>N</i>
NCSBI	0.313	0.620	0.067	418	0.197	0.769	0.011	0.023	366
[4]	0.3294	0.6465	0.0243	577	0.2316	0.7684	0.0	...	177
[5]	0.337	0.632	0.031	1239	0.222	0.767	0.009	...	718
[6]	0.3350	0.6054	0.0596	503	0.2341	0.7421	0.0109	0.0129	504
[7]	0.332	0.630	0.038	4850	0.217	0.776	0.008	...	875
[8]	0.327	0.612	0.061	1044	0.223	0.771	0.006	...	827
PGM									
	<i>PGM</i> <sup>1</sup>	<i>PGM</i> <sup>2</sup>	<i>N</i>		<i>PGM</i> <sup>1</sup>	<i>PGM</i> <sup>2</sup>	<i>N</i>		
NCSBI	0.774	0.226	833		0.805	0.195	777		
[4]	0.7435	0.2565	577		0.8305	0.1695	177		
[5]	0.751	0.248	1253		0.821	0.181	714		
[6]	0.7505	0.2495	503		0.7897	0.2103	504		
[7]	0.768	0.232	5972		0.812	0.188	1024		
[8]	0.771	0.229	1067		0.816	0.184	795		
Hp									
	<i>Hp</i> <sup>1</sup>	<i>Hp</i> <sup>2</sup>	<i>N</i>		<i>Hp</i> <sup>1</sup>	<i>Hp</i> <sup>2</sup>	<i>Hp</i> <sup>2M</sup>	<i>N</i>	
NCSBI	0.414	0.586	395		0.536	0.464	...	317	
[4]	0.3983	0.6017	585		0.5341	0.4660	...	176	
[5]	0.380	0.620	1263		0.535	0.454	...	721	
[6]	0.3489	0.6491	503		0.5558	0.3391	0.0981	504	
[7]	0.392	0.608	274		0.567	0.433	...	124	
[8]	0.422	0.578	860		0.550	0.450	...	460	
EsD									
	<i>EsD</i> <sup>1</sup>	<i>EsD</i> <sup>2</sup>	<i>N</i>		<i>EsD</i> <sup>1</sup>	<i>EsD</i> <sup>2</sup>	<i>N</i>		
NCSBI	0.898	0.102	388		0.931	0.069	343		
[4]	0.9064	0.0936	577		0.9350	0.0649	177		
[6]	0.8847	0.1153	503		0.9167	0.0833	504		
[7]	0.892	0.108	5377		0.916	0.084	973		
[8]	0.884	0.116	1025		0.904	0.096	770		
GLO									
	<i>GLO</i> <sup>1</sup>	<i>GLO</i> <sup>2</sup>	<i>N</i>		<i>GLO</i> <sup>1</sup>	<i>GLO</i> <sup>2</sup>	<i>N</i>		
NCSBI	0.430	0.570	309		0.297	0.703	327		
[6]	0.4573	0.5427	503		0.3591	0.6409	504		
[8]	0.436	0.564	313		0.327	0.673	310		
AK									
	<i>AK</i> <sup>1</sup>	<i>AK</i> <sup>2</sup>	<i>N</i>		<i>AK</i> <sup>1</sup>	<i>AK</i> <sup>2</sup>	<i>N</i>		
NCSBI	0.9665	0.0335	442		0.989	0.011	400		
[4]	0.9621	0.0379	580		0.9828	0.0173	174		
[6]	0.9712	0.0288	503		0.9970	0.0030	504		
[7]	0.963	0.037	5969		0.992	0.008	965		
[8]	0.962	0.038	1021		0.991	0.009	735		

TABLE 7—Continued.

Reference	Whites			Blacks				
	ADA							
	ADA <sup>1</sup>	ADA <sup>2</sup>	N	ADA <sup>1</sup>	ADA <sup>2</sup>	N		
NCSBI	0.946	0.054	436	0.995	0.005	399		
[6]	0.9423	0.0577	503	0.9921	0.0079	504		
[7]	0.949	0.051	5883	0.989	0.011	927		
[8]	0.952	0.048	1004	0.982	0.018	726		
6PGD								
	6PGD <sup>A</sup>	6PGD <sup>C</sup>	N	6PGD <sup>A</sup>	6PGD <sup>C</sup>	N		
NCSBI	0.9795	0.0190	317	0.9545	0.0440	309		
[6]	0.9781	0.0219	503	0.9554	0.0427	504		
[7]	0.981	0.019	4472	0.964	0.036	787		
[8]	0.983	0.017	950	0.955	0.045	826		
CA								
	CA <sup>1</sup>	CA <sup>2</sup>	N	CA <sup>1</sup>	CA <sup>2</sup>	N		
NCSBI	0.9985	0.0015	335	0.8885	0.1115	395		
[6]	1.00	0.0	503	0.9137	0.0863	504		
PepA								
	PepA <sup>1</sup>	PepA <sup>2</sup>	PepA <sup>8</sup>	N	PepA <sup>1</sup>	PepA <sup>2</sup>	PepA <sup>8</sup>	N
NCSBI	0.9985	0.0015	...	315	0.9225	0.0760	...	370
[6]	0.8820	0.0	0.1180	503	0.8688	0.0675	0.0637	504
[8]	1.00	0.0	...	300	0.948	0.052	...	488
Hb								
	Hb <sup>A</sup>	Hb <sup>S</sup>	N	Hb <sup>A</sup>	Hb <sup>S</sup>	Hb <sup>C</sup>	N	
NCSBI	1.00	...	365	0.9560	0.0345	0.0095	364	
[6]	1.00	...	503	0.9474	0.0367	0.0149	504	
[7]	0.999	0.001	6004	0.956	0.044	...	1025	
[8]	1.00	...	1040	0.953	0.035	0.012	789	

<sup>a</sup>Gene frequencies in the PGM subtyping system were not available from these references for comparative purposes.

TABLE 8—Lewis phenotype frequencies from three different populations.

Lewis Type	Observed Frequencies from North Carolina		Observed Frequencies from Delaware Valley [21]		Observed Frequencies from New York [22]	
	White, %	Black, %	White, %	Black, %	White, %	Black, %
Lewis a-b+	67.7	50.9	70.2	51.9	71.5	57.7
Lewis a+b-	20.4	20.4	20.1	19.6	22.8	23.1
Lewis a-b-	11.9	28.7	9.7	28.5	5.7	19.2
Individuals tested	548	432	935	833	460	411

rently use will allow NCSBI serologists for the first time to present population frequency data in court specific for our jurisdiction and that we have collected ourselves. We hope that this will eliminate many of the appeals and objections to this type of testimony in court.

So that other laboratories that have microcomputers may benefit from our experience, the author will make a copy of this program and a printout available to forensic science laboratories that make a request on official letterhead and send a blank 5¼-in. floppy disk. It is suggested that this program be used as a model for interested laboratories to set up their own programs suited to their special needs.

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### References

- [1] *State of North Carolina v. Larry Donnell Kirby*, N.C. Super. Ct. 80CRS9350, N.C. Ct. App. 8118SC877.
- [2] Tanton, R. L., "Computer-Assisted Calculation of Blood Frequency Data," presented at the Florida Serologist's and Microanalyst's Meeting, Palm Beach, FL, June 1979.
- [3] Wist, A. A., "Microcomputers and Serology," presented at the 1980 Combined Meeting in Louisville, KY of the Northeast, Midatlantic, Mideastern, and Southern Associations of Forensic Scientists, Auburn, AL, May 1980.
- [4] *Biochemical and Serological Methods in Bloodstain Analysis School Manual*, Federal Bureau of Investigation Academy, Quantico, VA.
- [5] Hagins, A. M., Shaler, R. C., Mortimer, C. E., Stuver, W. C., and Neilson, D. M., "Population Frequencies of Forensically Important Genetic Markers: Phosphoglucomutase, Erythrocyte Acid Phosphatase, and Haptoglobin," *Journal of Forensic Sciences*, Vol. 23, No. 3, July 1978, pp. 563-569.
- [6] Stolorow, M. D., Housel, D. L., Schaefer, J. R., Schoonover, J. L., Hauncher, J. D., Metzger, D. A., and Backos, G. B., "Blood Genetic Marker Study Greater Detroit Metropolitan Area: A Preliminary Report," presented at the 31st Annual Meeting in Atlanta, GA of the American Academy of Forensic Sciences, Colorado Springs, CO, Feb. 1979.
- [7] Grunbaum, B. W., Selvin, S., Pace, N., and Black, D. M., "Frequency Distribution and Discrimination Probability of Twelve Protein Genetic Variants in Human Blood as Functions of Race, Sex, and Age," *Journal of Forensic Sciences*, Vol. 23, No. 3, July 1978, pp. 577-587.
- [8] Grunbaum, B. W., Selvin, S., Myhre, R. A., and Pace, N., "Distribution of Gene Frequencies and Discrimination Probabilities for 22 Human Blood Genetic Systems in Four Racial Groups," *Journal of Forensic Sciences*, Vol. 25, No. 2, April 1980, pp. 428-444.
- [9] Mudd, J. L., "A Capillary Tube Method for the Lewis Typing of Red Blood Cells," *Journal of Forensic Sciences*, Vol. 28, No. 1, Jan. 1983, pp. 231-234.
- [10] *Biology Methods Manual*, Metropolitan Police Forensic Science Laboratory, London, England, 1978, pp. 2-51-2-54 and 2-67-2-71.
- [11] Nelson, M. S., "Detection of the Rare PGM<sub>1</sub><sup>3</sup> Allele: Further Biochemical and Genetic Characterization of the PGM<sub>1</sub><sup>3</sup> Isozymes," *Journal of Forensic Sciences*, Vol. 26, No. 1, Jan. 1981, pp. 75-81.
- [12] Wraxall, B. G., Bordeau, J., Harmor, G. C., and Walsh, J., "Final Report of the Blood Stain Analysis System," Contract J-LEAA-025-73, Law Enforcement Assistant Administration, Washington, DC, 1978.
- [13] "Sickle Cell Hemoglobinopathies," Procedure 15, June 1976, Helena Laboratories, Beaumont, TX.
- [14] Harmor, G., "Multisystem Approach to Red Cell Black Population Markers: Group IV," presented at the Spring Meeting in Savannah, GA of the Southern Association of Forensic Scientists Auburn, AL, May 1982.
- [15] Nelson, M. S., "Biochemical and Genetic Characterization of the Lowell Variant. A New Phenotype of 6-Phosphogluconate Dehydrogenase," *Human Genetics*, Vol. 62, No. 4, Dec. 1982, pp. 333-336.

- [16] "Instruction LKB Ampholine PAG plates pH 5.0-6.5 for Phenotyping of Phosphoglucomutase Isoenzymes," *LKB Technical Information Manual I-1804-121-E02*, LKB, Bromma, Sweden.
- [17] Strickberger, M. W., "Probability and Statistical Testing," *Genetics*, MacMillan, New York, 1968, pp. 126-152.
- [18] Giblett, E. R., "Haptoglobin," *Genetic Markers in Human Blood*, Blackwell Scientific Publications, Oxford, England, 1969, pp. 63-125.
- [19] Kirk, R. L., "The Problem of Hp2-1 (Mod) and Hp 0," *Haptoglobin Groups in Man, Monographs in Human Genetics*, Vol. 4, L. Beckman and M. Hauge, Eds., S. Karger, Basel, Switzerland, 1968, pp. 31-39.
- [20] Dykes, D. D. and Polesky, H. F., "Comparison of Rare PGM<sub>1</sub> Variants by Isoelectric Focusing and Conventional Electrophoresis: Identification of Five New Variants," *Electrophoresis*, Vol. 2, No. 5-6, Dec. 1981, pp. 323-326.
- [21] Molthan, L., "Lewis Phenotypes of American Caucasians, American Negroes, and Their Children," *Vox Sanguinis*, Vol. 39, No. 6, Dec. 1980, pp. 327-330.
- [22] Miller, E. B., Rosenfield, R. E., Vogel, P., Harber, G., and Gibbel, N., "The Lewis Blood Factors in American Negroes," *American Journal of Physical Anthropology*, Vol. 12, No. 3, Sept. 1954, pp. 427-443.
- [23] Zmijewski, C. M. and Fletcher, J. L., "Lewis, Lutheran, and Secretion," *Immunohematology*, 2nd ed., Appleton-Century-Crofts, New York, 1972, pp. 84-98.

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