

BLOOD GROUPS OF THE *ABO*, *MN* AND *CDE-cde* SYSTEMS IN
THE NATIVE POPULATIONS OF RUANDA-URUNDI
TERRITORIES*

BY P. O. HUBINONT, M.D., J. HIERNAUX, M.D. AND TH. MASSART-GUIOT

*Blood Group Laboratory and Hygiene Laboratory (Université Libre de Bruxelles) and
Research Centre of the Ruanda-Urundi Territories (I.R.S.A.C. Astrida)*

In the Ruanda and Urundi territories the population may be divided into three well-defined stocks:

(1) The *Batwa* have a typically pygmoid physique: small in height, with a high cormic index,† they have a short and broad nose and a very marked pilosity. However, they are very different from the *Bambutu* Pygmies of the Ituri forest, these differences being partly due to the mixing of the former with Bantus. They live in small groups, in the forest, hunting and gathering food. Some of them have left the forest and live in the villages, where they work as potters or as dancers in the 'intore' troops for the entertainment of kings or great chiefs. They form a very small minority and have not been considered in the present study.

(2) The greater part of the population consists of *Bahutu* who are agricultural and pastoral Bantus. A long cohabitation with populations of 'Hamitic' stock may explain the frequency of non-negroid anthropological characters as a high and thin nose, but they do not differ much from other Bantus possessing the attenuated negro face, as the South-African Bantus for instance. They live under the power of the *Batutsi*, in a state of bondage which is similar to that of the Middle Ages in Europe but which is steadily being modified under European influence.

(3) The *Batutsi* constitute about one-tenth of the population. They entered the country a few centuries ago and have ruled it ever since, dominating the *Bahutu* easily by their intelligence and their physical prestige, with little use of force. Tall (176 cm. as a mean), thin, long-legged, they have a long head, a high and narrow nose (mean nasal index of 70), an orthognathic face. Anthropometrically speaking they differ greatly from the negroes in spite of their skin colour and woolly hair.

The political frontier between Ruanda and Urundi does not appear to have constituted an ethnical barrier: on both sides are found the same groups, the same speech, the same tradition and culture. It therefore seems logical to collect in one sample the homologous groups of both countries. However, two restrictions must be taken into consideration:

(1) In the south-west and south-east of the Urundi, that is, in the *Imbo* and *Mosso* regions, are found populations which differ anthropometrically from the *Bahutu*. They have not been taken into account in the present article, but they will be studied later.

(2) In the Urundi the *Batutsi* may be anthropometrically divided in two groups:

(a) the *Batutsi* living in the pastoral regions, upon the high plateau of the Congo-Nile watershed, or in the *Bugessera*, being in a majority, and never mixing with Bantus;

(b) the *Batutsi* of the rest of Urundi, where they are in a small minority, are mixed with the *Bahutu* and have anthropological features intermediate between the extreme types of the two races.

* Aided by a grant from the Royal Institute for Scientific Research in Central Africa (I.R.S.A.C.).

† Sitting height/stature \times 100.

As we shall see, the distribution of the blood groups is in perfect accordance with the anthropometric data. These will be published elsewhere by one of us (Hiernaux, 1953).

These preliminary notions being given, we feel that we may pass on to the subject of the present communication.

A. METHODS

(1) *Taking and transport of the samples*

Blood was taken by venepuncture in 'venules'. These were packed in thermally insulated containers and surrounded with melting ice. An even temperature was maintained during the whole journey by adding ice whenever necessary.

The samples were studied as a rule within 48 hr. of taking. In certain cases there was a delay, which, however, never exceeded 4 days, between the taking and the completion of the serological work. The serological reactions were always clear-cut and never gave the impression that agglutinogens had deteriorated.

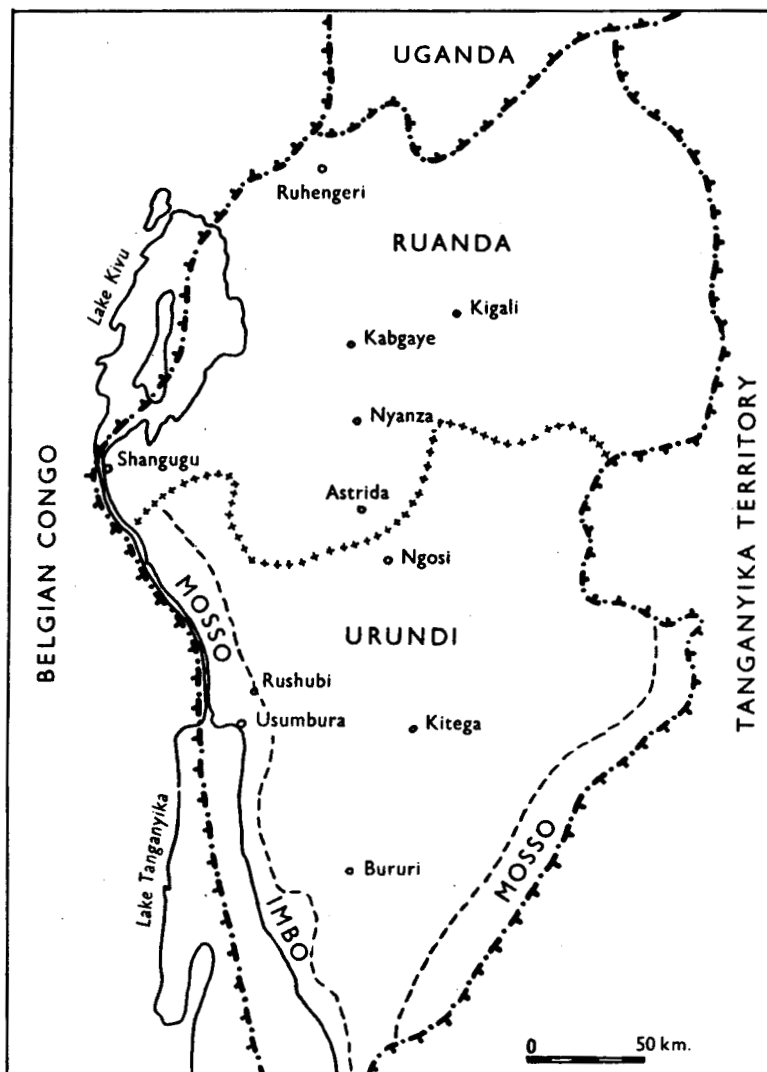


Fig. 1. Map showing the Ruanda-Urundi territories.

The *Bahutu* samples were taken in Kabgaye, Astrida, Nyanza, Kigali, Ruhengeri and Shangugu in the Ruanda, and Ngosi, Kitega, Rushubi and Usumbura in the Urundi.

The *Batutsi* samples came from Kabgaye, Astrida, Nyanza, Kigali, Ruhengeri and Shangugu in the Ruanda, and Bururi in the Urundi.

The 'half-bred' *Batutsi* of the Urundi were found in Ngosi, Kitega, Rushubi and Usumbura.

The location of these places will be found on the annexed map (Fig.1). Samples were taken from widely separated places in order to get a close agreement with the general population. Close consanguinity has always been avoided.

(2) Serological technique

We have used for blood-group determination and for *Rh* genotyping, the techniques described by Race & Sanger (1950) which we had learned some years ago in their laboratory.

As stated above, all the reactions were clear-cut and never needed discussion. The negatives were controlled microscopically after centrifugation at low speed for one minute.

(3) Statistical methods

We have made a large use of statistical methods applied to gene frequencies, which we have found very useful.

B. RESULTS

We had classified the samples, as they reached the laboratory, into four groups according as they came from Ruanda or Urundi or had been taken in Bahutu or Batutsi. The result of this classification is set out in full in Table 1.

These four groups had been compared by a simple χ^2 , applied to the *ABO*, *MN* and *CDE-cde* distributions. The χ^2 values and figures for *p* in each case were found to be:

Compared groups	Blood group system	χ^2	<i>n</i>	<i>p</i>
Ruanda-Bahutu v. Batutsi	<i>ABO</i>	16.7834	5	<0.01
	<i>MN</i>	4.1654	2	0.10-0.05
	<i>CDE-cde</i>	2.0757	5	0.90-0.80
Urundi-Bahutu v. Batutsi	<i>ABO</i>	6.1225	5	0.30-0.20
	<i>MN</i>	0.8017	2	0.70-0.50
	<i>CDE-cde</i>	3.7778	5	0.50-0.30
Batutsi-Ruanda v. Urundi	<i>ABO</i>	2.7906	5	0.80-0.70
	<i>MN</i>	0.8017	2	0.70-0.50
	<i>CDE-cde</i>	4.5349	5	0.50-0.30
Bahutu-Ruanda v. Urundi	<i>ABO</i>	3.1104	5	0.70-0.50
	<i>MN</i>	0.5831	2	0.50-0.30
	<i>CDE-cde</i>	6.4833	5	0.30-0.20

The first conclusion drawn from these results was that if *ABO* blood-group distributions were significantly different between Batutsi and Bahutu in the Ruanda, the same situation was not found in the Urundi. We could find two possible reasons for this apparent discrepancy with the

anthropological data: first that our samples were too small, second that there might be some kind of crossing of certain Batutsi of the Urundi with the Bahutu population.

For these reasons we have constituted three groups on *purely anthropometrical grounds*.

The first group is made of the whole lot of Batutsi of the Ruanda plus twenty-two samples taken in the Urundi in a region (see above) where they do not mix with Bahutu.

Table 1. *Observed phenotype frequencies when classified according to the geographical origin of the samples*

	A_1	A_2	B	O	A_1B	A_2B	M	N	MN	R_0	$R\frac{y}{z}$	R_1	R_2	r	R_1R_2	R_1R_1	R_1^w	R'	R'^w	$R\frac{y}{z}$	Total	
Ruanda																						
Batutsi	11	11	10	71	1	1	40	21	44	66	4	12	9	6	2	2	3	—	1	—	—	105
Bahutu	29	9	33	72	4	1	39	39	70	97	10	12	14	7	2	2	3	1	—	—	—	148
Urundi																						
Batutsi	11	8	7	44	2	—	24	13	35	39	5	15	6	3	2	1	—	—	—	—	1	72
Bahutu	25	11	24	57	2	—	36	29	54	85	2	15	10	3	3	—	—	1	—	—	—	119
Total																						444

Table 2. *Observed phenotype frequencies when classified upon anthropometric grounds*

	A_1	A_2	B	O	A_1B	A_2B	M	N	MN	R_0	$R\frac{y}{z}$	R_1	R_2	r	R_1R_2	R_1R_1	R_1^w	R'	R'^w	$R\frac{y}{z}$	Total	
Batutsi	15	13	12	84	2	1	51	24	52	79	7	15	12	6	2	2	3	—	1	—	—	127
'Half-bred'	7	6	5	31	1	—	13	10	27	26	2	12	3	3	2	1	—	—	—	—	1	50
Batutsi																						
Bahutu	54	20	57	129	6	1	75	68	124	182	12	27	24	10	5	2	3	2	—	—	—	267
Total																						444

The second group contains the rest of the Urundi Batutsi which were known to be mixed with Bahutu.

In the third group have been placed all the Bahutu.

The actual distribution of different groups has been written down in Table 2.

Comparison of these three groups by the χ^2 test gave the following figures:

Compared groups	Blood group system	χ^2	n	p
Bahutu-Batutsi	ABO	15.9152	5	<0.01
	MN	5.9602	2	<0.05
	$CDE-cde$	1.6545	5	0.90-0.80
Batutsi-'half-bred' Batutsi	ABO	4.6681	5	0.50-0.30
	MN	2.6877	2	0.30-0.20
	$CDE-cde$	6.0883	5	~0.30
Bahutu-'half-bred' Batutsi	ABO	6.4266	5	0.30-0.20
	MN	1.0834	2	0.70-0.50
	$CDE-cde$	10.2932	5	0.10-0.05

This second way of sampling gives more significant differences between Bahutu and Batutsi. Curiously enough there is no significant difference for the $CDE-cde$ blood groups.

C. GENE FREQUENCIES

Gene frequencies have been calculated according to the classical methods described in many specialized treatises. We have referred mainly to Race & Sanger (1950) (Tables 3, 4, 5).

Table 3. *Frequencies of ABO blood groups in Batutsi*

Phenotypes	Observed figures		Calculated figures	
	No.	%	%	No.
A_1	15	11·8111	11·9132	15·1297
A_2	13	10·2362	10·3571	13·1535
B	12	9·4488	9·5513	12·1302
O	84	66·1417	66·7489	84·7712
A_1B	2	1·5748	0·7390	0·9385
A_2B	1	0·7874	0·6904	0·8768
	127	100·0000	99·9999	126·9999

$\chi^2 = 1·4238$ for four degrees of freedom.

Table 4. *Frequencies of MN blood groups in Batutsi*

Phenotypes	Observed figures		Calculated figures	
	No.	%	%	No.
M	51	40·1575	36·7599	46·6851
N	24	18·8976	15·5000	19·6850
MN	52	40·9449	47·7401	60·6299
	127	100·0000	100·0000	127·0000

$\chi^2 = 2·5730$ for one degree of freedom.

Table 5. *Frequencies of CDE-cde blood groups in Batutsi*

Phenotypes	Observed figures		Calculated figures	
	No.	%	%	No.
$- + - + - * = R_0$	86	67·7165	67·7108	85·9927
$- (+) - + - = R_0^w$				
$+ + - + - = R_1$	18	14·1732	14·7029	18·6727
$- + - + + = R_1^w$				
$- - - + - = r$	6	4·7245	4·7263	6·0024
$+ + + + - = R_1R_2$	2	1·5748	1·0455	1·3278
$+ + - - - = R_1R_1$	2	1·5748	0·7981	1·0136
$- - - + + = R'^w$	1	0·7874	1·0433	1·3250
$- + + + - = R_2$	12	9·4488	9·9730	12·6657
	127	100·0000	99·9999	126·9999

$\chi^2 = 1·4494$ for five degrees of freedom.

* Reactions with C, D, E, c and C^w anti-sera.

The calculations for the ABO system (p_1, p_2, q and r) were established following Wellisch & Thomsen (1930). For the m and n genes we used Wiener & Vaisberg's (1931) formula:

$$m = M + \frac{MN}{2}, \text{ etc.}$$

For the *CDE-cde* gene calculations, we made use of the simpler method described by Fisher (Race *et al.* 1946). However, we have slightly altered the calculation of *CDe*. It is classical to use the expression

$$CDe = \sqrt{\{(++--)+(+-)\}} - \text{frequency of } CDe.$$

As *CDe* is usually rare in these populations we felt that the observed frequency of phenotype *++--* might differ widely from its actual frequency, and we used the expression

$$CDe = \sqrt{\{(++--)+(++-+)+(-+-+)+(+-+)+(- - -+)\}} - \text{frequencies of } cDe, Cde \text{ and } cde,$$

which had the advantage of being based on a larger sample.

On the other hand, for simplification we considered as *C* the elementary gene *C^w*, and as *D* the character *D^u*. However, these characters were found in a certain number of cases. This is certainly an important fact to point out, especially as regards the *D^u* character. We had already drawn attention in a previous work (Hubinont, 1949) to the fact that ignoring it would lead to rather frequent errors when testing Africans. Among a total of 444 determinations, *D^u* was found 21 times in the *cD^ue* combination and once in *cD^uE*, that is, in 5% of all determinations.

Table 6. *Frequencies of ABO blood groups in Bahutu*

Phenotypes	Observed figures		Calculated figures	
	No.	%	%	No.
<i>A</i> ₁	54	20.2247	19.7686	52.7822
<i>A</i> ₂	20	7.4906	7.3286	19.5674
<i>B</i>	57	21.3483	20.8546	55.6818
<i>O</i>	129	48.3146	47.2244	126.0891
<i>A</i> ₁ <i>B</i>	6	2.2472	3.4061	9.0943
<i>A</i> ₂ <i>B</i>	1	0.3746	1.4176	3.7850
	267	100.0000	99.9999	266.9998

$\chi^2 = 3.2343$ for four degrees of freedom.

Table 7. *Frequencies of MN blood groups in Bahutu*

Phenotypes	Observed figures		Calculated figures	
	No.	%	%	No.
<i>M</i>	75	28.0899	26.3282	70.2963
<i>N</i>	68	25.4682	23.7062	63.2955
<i>MN</i>	124	46.4419	49.9656	133.4082
	267	100.0000	100.0000	267.0000

$\chi^2 = 1.0756$ for one degree of freedom.

Table 8. *Frequencies of CDE-cde blood groups in Bahutu*

Phenotypes	Observed figures		Calculated figures	
	No.	%	%	No.
- + - + - = R_0	194	72·6592	72·6958	194·0978
- (+) - + - = R_0u				
+ + - + - = R_1				
- + - + + = R_1w				
- - - + - = r	10	3·7453	3·7442	9·9970
+ + + + - = R_1R_2	5	1·8726	0·7882	2·1045
+ + - - - = R_1R_1	2	0·7491	0·3997	1·0672
+ - - + - = R'	2	0·7491	0·7544	2·0142
- + + + - = R_2	24	8·9888	10·8142	28·8739
	267	100·0000	100·0000	266·9999

$\chi^2 = 5·6919$ for five degrees of freedom.

Table 9. *Gene frequencies for the ABO, MN and CDE-cde blood groups systems (%)*

	Batutsi	Bahutu
$A_1 (p_1)$	6·54	12·35
$A_2 (p_2)$	6·11	5·14
$B (q)$	5·65	13·79
$O (r)$	81·70	68·72
M	60·63	51·31
N	39·37	48·69
$cDe (R_0)$ including cD^ue	63·37	68·08
$CDe (R_1)$ including C^wDe	6·94	4·73
$cDE (R_2)$ including cD^wE	5·67	5·98
$Cde (R')$ including C^wde	2·28	1·86
$cde (r)$	21·74	19·35

The observed and calculated frequencies of blood groups, and the statistical tests controlling their accuracy, are given in Tables 3, 4, 5, 6, 7, 8 and 9. They show no important discrepancies between calculated and observed figures. The worst results were found in the *MN* distribution where we consider that the serological technique was not as good as for the *ABO* and *CDE-cde* blood groups.

However, we may admit that the observed distributions are satisfactory and that the frequencies attributed to the genes have been verified by experiment.

CONCLUSION

What have we learned from the blood groups upon the racial position of Batutsi and Bahutu in Africa?

(a) *The ABO system*

Gene frequencies for p , q and r link the Bahutu together with other 'negroes', especially with the 'Bantus' of the Belgian Congo.

The Batutsi, on the other hand, are placed close to certain tribes of South Africa (for instance, the Inyambane of Mozambique, or the Tchopi) and even closer to some Oriental and North Africans as some Erythrean natives, the Tebu of the Tibesti region, some stocks of Shluh, etc. They exhibit also certain similarities to Samaritan Jews, these affinities fitting perfectly with the 'non-negroid' traits revealed by the anthropometrical analysis. If we indicate by triangular

co-ordinates the relative position of different Africans, including the populations studied in the present investigation, we notice (Fig. 2) that between the representative points of Batutsi and Bahutu are found, approximately on a straight line, more southern populations as Tchopi, Inyambane, Swazi, Sotho, South African Bantus and Zulus.* This leads us to consider that all these populations might have a common Bantu ground upon which would be

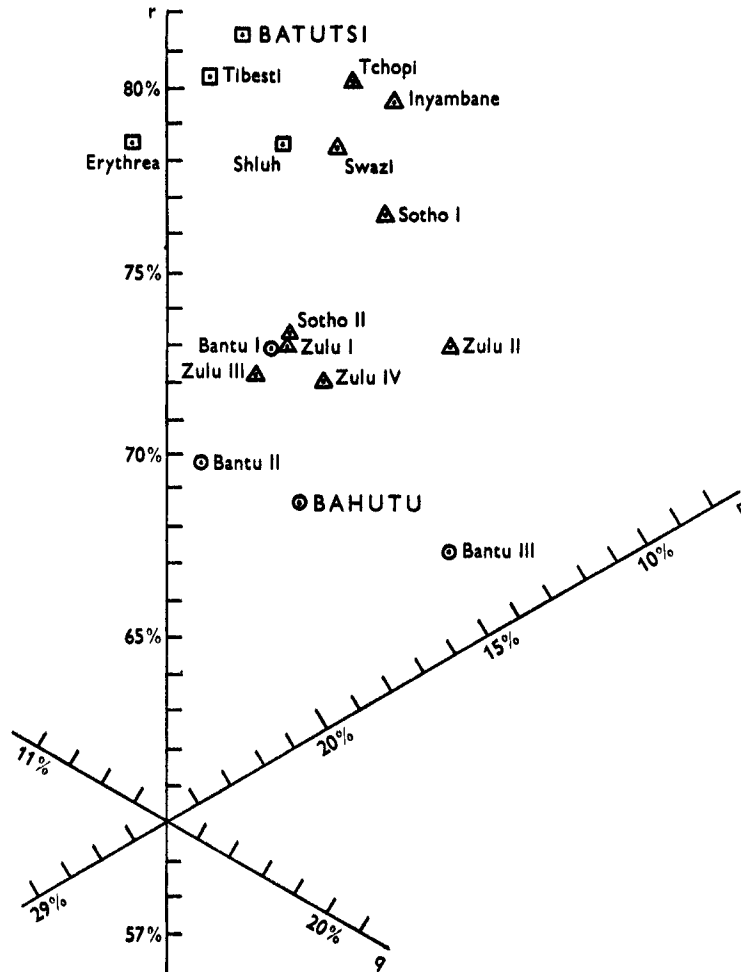


Fig. 2. Location of Batutsi and Bahutu together with other African populations in Streng's triangular co-ordinates according to the frequencies p , q , r of the genes A , B , O respectively.

grafted a variable 'Hamitic' contribution. Here also the serological results confirm the observations made by physical anthropologists who have noted the high frequency of non-negroid facies among South African Bantus and of ethnologists who find, in the same populations, the sometimes highly marked influence of the Hamitic pastoral culture.

(b) *The A_2/A_1 coefficient*

The proportion between A_2 and A_1 is 0.86 in Batutsi and 0.37 in Bahutu. The observed ratio among the latter is similar to that found in North American Negroes (Boyd, 1950).

* The gene frequencies were recorded in Boyd (1939).

On the contrary, the proportion found in Batutsi is by far the highest among the populations of the world, so far described, other than the Lapps (Allison, Hartmann, Brendemoen & Mourant, 1952).

(c) *The MN system*

The gene frequencies for the *MN* system are very different between Batutsi and Bahutu and may distinguish the two races.

(d) *The CDE-cde system*

While *ABO* and *MN* systems could separate sharply the Batutsi and the Bahutu, the *CDE-cde* system does not have any significantly different distribution. This has already been noticed for other African and non-African populations. It is, of course, a puzzling observation which we have not been able to explain reasonably.

We feel that any attempt to do so would be superfluous, and that further sero-anthropological studies will probably lead to its solution.

However, we might admit that the present gene-frequency distribution for the *Rh* system in the Batutsi is remote from the original state, prior to their isolation. Among the factors modifying gene frequencies in isolates, account cannot be taken of the *random genetic drift* as defined by Sewall Wright, for this affects only genes of lower frequencies. If we consider that the most frequent *Rh* gene group is the *cDe* which is generally considered as a negro character, and if we revert to the fact that besides 'Hamitic' traits the Batutsi possess woolly hair and a very dark skin which are truly negro, we should be inclined to favour the hypothesis of an association between the woolly hair, the colour of the skin, and a high *cDe* frequency.

We should then consider that *natural selection*, having an influence upon the physical characters had also altered either indirectly or directly the gene frequencies for the *CDE-cde* blood-group system.

We wish to thank Dr J. J. Van Loghem, Jr., to whom we are indebted for the generous gift of anti-*Rh* test sera, to the Administrative Services of the S.A.B.E.N.A. (Belgian airlines) for the valuable help in transportation problems, and to the Executive Committee of the I.R.S.A.C. for the outstanding financial and moral support given to our work during the last three years.

Dr A. E. Mourant has also contributed to its success by the gift of test sera, but mainly by his constant and friendly attention and by his kind and comprehensive criticisms in the writing of the present paper.

REFERENCES

- ALLISON, A. C., HARTMANN, O., BRENDEMOEN, O. J. & MOURANT, A. E. (1952). The blood groups of Norwegian Lapps. *Acta path. microbiol. scand.* **31**, 334.
- BOYD, W. C. (1939). *Tabul. biol., Den Haag*, **17**, 113.
- BOYD, W. C. (1950). *Genetics and the Races of Man*. Boston: Little and Brown.
- HIERNAUX, J. (1953). Les Caractères physiques des Populations du Ruanda-Urundi (to be published).
- HUBINONT, P. O. (1949). Importance de la variété *D^u* de l'antigène *D* dans les recherches relatives au système *Rh-Hr*. *C.R. Soc. Biol., Paris*, **143**, 581.
- RACE, R. R., MOURANT, A. E. & MCFARLANE, MARJORY N. (1946). Travaux récents sur les antigènes et anticorps *Rh* avec une étude particulière de la théorie de Fisher. *Rev. Hémat.* **1**, 9.
- RACE, R. R. & SANGER, R. (1950). *Blood Groups in Man*. Oxford: Blackwell.
- WELLISCH, S. & THOMSEN, O. (1930) Über die vier-gen-hypothese Thomsens. *Hereditas, Lund*, **14**, 50.
- WIENER, A. S. & VAISBERG, M. (1931). Heredity of the agglutinogens *M* and *N* of Landsteiner & Levine. *J. Immunol.* **20**, 371.