

ON THE GENETICS OF BETANIN EXCRETION

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THE ability to excrete beetroot red pigment (betanin) in the urine in 11 English families has been reported by Allison and McWhirter¹. When both parents were excretors (two families), only excretor children were found, which suggested recessive inheritance. Saldanha *et al.*² found that the frequencies of excretor individuals showed significant race heterogeneity. The trait probably illustrates another case of polymorphism in inborn patterns of human metabolism. However, before the betanin excretion can profitably be used in racial and linkage investigations, its inheritance must be more accurately known. In this paper some family data on the betanin excretion are presented and analyzed.

Materials and Methods

During a genetical and anthropological investigation carried on in a Dutch colony in the State of São Paulo, Brazil³, betanin excretion was studied in 18 families, which included 111 children. The individuals were tested by means of a procedure described by Saldanha *et al.*² About 100 g. of cooked beetroot were given to each individual. Two urine samples were collected in test tubes, one before and the other five hours after the ingestion of the beetroot. By comparing visually the two samples, subjects were classified as non-excretor—(—, no visible trace of pigment) or excretor, with three different categories: (+, very weak tint); (++, medium intensity of red color); (+++, strong red color). Instructions were given to all parents to make sure their children would actually eat their share of the root. Most families in Table I were tested in October, 1959. One child in Family 16, the mother and one child in Family 17 and Families 7,

11, 15 and 18 were tested in May, 1960. The parents and four children in Family 2 were retested in May, 1960 and had their excretion types confirmed, with the exception of one child who passed from minus to plus.

Results and Discussion

The distribution of the children according to sex and excretion type was:

	—	+	++	+++	Total
Male	32	14	9	6	61
Female	29	6	10	5	50

No significant sex difference is apparent: $\chi^2_{(3)} = 2.4$; $P = >0.45$.

Table I presents the frequency of children in each excretion type for each of the families studied. The pooled frequencies for each mating type are shown in Table II. The distribution of phenotypes among children is significantly depend-

TABLE I. Betanin excretion in 18 Dutch families in Brazil*

Family	Parents		Children			
	♂	♀	—	+	++	+++
1	—	—	6	1		
2	—	—	6	1		
3	—	—	6	1		
4	—	—	7	1		
5	—	—	5		1	
6	—	—	3		2	
7	—	—	1		4	2
8	—	—	4	2	1	1
9	+	—	3	1		
10	—	+	4	1	2	
11	—	+	3	1		1
12	+	—	3		3	1
13	+	—	1	1	1	1
14	+	+		3		
15	+	+	1	4	3	
16	+	+	2	2	1	2
17	+++	—	5	1	1	2
18	++	+++	1			3
Total			61	20	19	11

*— = lack of excretion; + = very small excretion; ++ = medium excretion; +++ = strong excretion.

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ent on the type of mating ($\chi^2_{(12)} = 41.3$; $P < 0.0001$).

As shown in Table I, three of four families where both parents were excretors produced four non-excretor children. This result disagrees with the hypothesis of simple recessive inheritance as suggested by Allison and McWhirter¹. It is unlikely that these cases are the result of mistaken interpretation or of illegitimacy. However, assuming

that this could be so, the recessive hypothesis can be tested by a consideration of the segregation ratios among the offspring born to the remaining mating types.

Assuming that the excretor phenotypes +, ++ and +++ are recessive homozygotes (*ee*) the genotype of each parent in the sample is directly known. Since each of the eight couples composed by non-excretor parents had one or more

TABLE II. Distribution of children's phenotypes according to the mating types of the parents (data from Table I)

Mating types	Number of families	Children				Total
		—	+	++	+++	
— × —	8	38	6	8	3	55
+ × —	5	14	4	6	3	27
+++ × —	1	5	1	1		7
+ × +	3	3	9	4	2	18
+++ × ++	1	1			3	4
Total	18	61	20	19	11	111

Heterogeneity: $\chi^2_{(12)} = 41.3$; $P < 0.0001$

TABLE III. Comparison between observed and expected numbers of excretor and non-excretor children born to different genotypic matings, assuming that +, ++ and +++ subjects are recessive homozygotes (see text)

Genotypic mating	Number of families	Children				Total	χ^2	P
		Non-excretor ($EE+Ee$)		Excretor (ee)				
		Obs.	Exp.	Obs.	Exp.			
$Ee \times Ee$	8	38	41.25	17	13.75	55	1.03	>0.50
$Ee \times ee$	6	19	17	15	17	34	0.48	>0.50
$ee \times ee$	4	4	0	18	22	22	---	---
Total	18	61		50		111	1.51	>0.50

TABLE IV. Comparison between observed and expected number of excretor and non-excretor children born to different phenotypic mates, assuming that only ++ and +++ subjects are recessive homozygotes (see text)

Mating types	Number of families	Children				Total	χ^2	P
		Non-excretor		Excretor				
		Obs.	Exp.	Obs.	Exp.			
Non-excretor $\times$ non-excretor	16	74	95.0	26	5.0	100	92.8	<0.0001
Non-excretor $\times$ excretor	1	6	5.4	1	1.6	7	0.3	>0.60
Excretor $\times$ excretor	1	1	(0)	3	(4)	4	—	—
Total	18	81		30		111	93.1	<0.0001

excretor children, they must be heterozygotes. Similarly since each of the six couples composed by excretor and non-excretor parents had at least one excretor child, the non-excretor parent in these couples should be heterozygotes. Table III shows a comparison between observed and expected numbers of excretor and non-excretor children born to the three different genotypic matings ($Ee \times Ee$; $Ee \times ee$; $ee \times ee$). Except for the four non-excretor children produced by the mating of two recessive homozygotes ($ee \times ee$), the result is in agreement with the recessive hypothesis ($\chi^2_{(2)} = 1.51$; $P = >0.50$).

It is not clear from Allison and McWhirter's paper whether subjects in our class (+, very weak tint) were considered by them as "excretors" (i.e., recessive homozygotes) or "non-excretors." Under the hypothesis that + phenotypes are non-excretors, then just one non-excretor child has been born to excretor parents (Table I, family 18), instead of four. However this hypothesis can be tested only indirectly. Among the 36 parents only three were ++ or ++++. Therefore under the present hypothesis the frequency of recessive homozygotes is $q^2 = 0.08$ and the gene frequency is $q = 0.29$. Starting from this gene frequency the expected numbers of "excretor" children (++ and ++++) born to "non-excretor $\times$ non-excretor" and "non-excretor $\times$ excretor" matings, were calculated and compared to the observed numbers (Table IV). The deviation is highly significant ($\chi^2_{(2)} = 93.1$; $P < 0.0001$). In spite of the sampling error in the evaluation of the frequency of recessive homozygotes, this seems to rule out the hypothesis.

The frequency of excretors is considerably greater among our Dutch sample (0.39) than among English (0.10) as

determined by Allison and McWhirter¹; but it is not much greater than that found among whites of Portuguese and Italian derivation in São Paulo, Brazil². If only phenotypes ++ and +++ are considered as "excretors", then the frequency among the Dutch drops to 0.08, very near to the value for the English sample. However, as we have seen (Table IV), the departure from the recessive hypothesis becomes great.

It is clear that discrepancies in the results of genetical analysis and population differences in the frequency of excretors can be, at least partly, attributed to differences in technique. Therefore, data obtained by means of a more refined and standardized technique should be relevant to the problem.

Summary

The ability to excrete beetroot pigment (betanin) in the urine was investigated in 18 Dutch families (36 parents and 111 children) living in the State of São Paulo, Brazil. Analysis of the data confirms that the trait is hereditary and would agree with a simple recessive scheme of inheritance, as previously suggested, if allowance is made for some exceptions found among the children. It is emphasized that discrepancies observed in the genetical analysis as well as differences in frequencies of excretors for betanin among populations can, at least partly, be attributed to the unreliability of present techniques of sampling.

Literature Cited

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