lamina area and upper lamina area/upper lamina dry weight  $(r_3 = -0.51, P = 0.09)$ . From the value of  $r_1$ it is evident that there was little association between the areas of the lower and upper laminæ. On the other hand,  $r_2$  and  $r_3$ , although of doubtful significance, suggested that the dry weight of the upper laminæ was to some extent dependent on the area of the lower laminæ and that the area per unit weight of the upper laminæ was greater in plants with small lower lamina area. Moreover, it is thought possible that closer correlations might have been obtained if both area and weight had been measured on the same plants rather than estimated separately from the two halves of a plot.

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#### MICROBIOLOGY

# Isolation of Diploid Strains of Dictyostelium discoideum from Haploid Populations

PLOIDAL composition is a clonally inherited property in D. discoideum<sup>1</sup>. Three types of strains exist: stable haploid, stable diploid, and metastable. In the last, each clone contains appreciable proportions of haploid and diploid cells indicating a rate of interconversion substantially higher than in the first two types. Each of these ploidal varieties can be isolated from the others2.

Two stable diploid stocks, heterozygotic for two pigment markers, have been isolated from mixed cultures of the respective mutant (metastable) strains, and these in turn have yielded, at very low frequency (10-3-10-2), stable haploid progeny which fell into the four expected segregant classes<sup>2</sup>. Several unmarked diploid stocks have also been isolated from cultures of the haploid wild-type. However, these represent rare events and the conditions for their isolation have not been clearly understood. The present communication describes a method by which diploid clones can be isolated from stable haploid cultures at relatively high frequency. The results indicate that the

individuals capable of yielding these diploid clones are to be found, after the cessation of growth, among the amoebae which fail to enter the cell aggregates and, thus, the fruiting bodies formed by the predomin-

antly haploid population.

A stable haploid white mutant of D. discoideum, Wh-1, was used in work recorded here. This ensured that the diploid strains derived from such cultures could not have been contaminants from diploid strains already at hand since all the latter form fruiting bodies possessing the wild-type (yellow) pigment. The stock was grown on SM agar<sup>3</sup> in association with Aerobacter aerogenes. The mature fruiting bodies that appeared were removed and the unaggregated amoebae that remained were used as the inoculum for the next passage. Two serial passages were accomplished in this manner. After each passage, unaggregated amoebae were isolated with a micromanipulator, placed on fresh agar, grown with A. aerogenes and the ploidal compositions of the resultant clones were determined<sup>1,2</sup>. Table 1 is a summary of the data. The normal incidence of cells capable of yielding diploid clones in stable haploid stocks cultivated

		Table 1			
Passage	Inoculum	No. of clones derived from unaggregated cells	Ploidal con Stable haploid	mposition Stable diploid	of the clones Metastable
I	Spores	62	62	0	0
II	Unaggregated cells	74	73	1	0
III	Unaggregated cells	76	62	9	5

estimated as  $<10^{-4}$  (ref. 2). In contrast, two serial passages using unaggregated amoebae as the inocula resulted in a frequency, among the unaggregated cells, of 18 per cent stable diploid and metastable strains. absence of selective markers for the isolation of heterozygotes, the method described here may be of value.

This work was supported by grant (G-12900) from the U.S. National Science Foundation.

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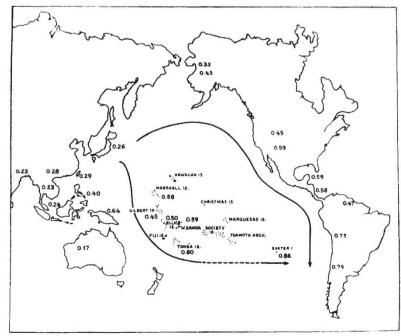
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#### **ANTHROPOLOGY**

## Haptoglobin Types in Inhabitants of Easter Island

SINCE the discovery by Smithies1 of the polymorphic character of haptoglobins a widespread interest has arisen in the distribution of haptoglobin, types among various ethnic groups. In this connexion Easter Island natives, who represent a maritime isolate in Eastern Polynesia, 2,000 miles from the coast of South America, are of particular interest.

The first contact of these inhabitants with the occidental world occurred in the eighteenth century when the total population of Easter Island is estimated to have been approximately 3,000. During the decade 1860-70, the population underwent a steady decrease as the result of Peruvian slave raids, smallpox epidemics and migration to Tahiti and Mangareva. Only about 200 or 300 natives remained in 1871. After 1871, a steady population increase has been observed, with the immigration of Euro-



from a spore inoculum (passage 1) has been Fig. 1. Hp1 gene frequency distribution in Pacific populations modified from E. Giblett (ref. 4)

							Table	1						
Desar	**	n	Total	$_{1-1}^a$	<i>b</i> 2-1	$^{c}_{2-2}$	2a+b	$\frac{2(a+b+c)}{y}$	w	wx	wy	$Hp^{1}$	S.E.	$\chi^2$
Pure Eastern Islanders	Unrelated Related	1 2 3 4	$^{6}_{12}$ $^{6}_{12}$	5 7 6 8	1 5 0 4	0 0 0 0	11 19 12 20	$12 \\ 24 \\ 12 \\ 24$	1 0.666 0.500 0.400	11 12·66 6·00 8·00 37·66	12 16.00 6.00 9.60 43.60	0.863	0.059	0.951
Admixed Eastern Islanders	Unrelated Related	$\begin{array}{c}1\\2\\3\\4\end{array}$	40 38 33 12	29 21 19 7	10 16 14 5	1 1 0 0	68 58 52 19	80 76 66 24	68 0.666 0.500 0.400	68 38·68 26·00 7·60 140·28	80 50·66 33·00 9·60 173·26	0.809	0.028	2.761
		$p\mathrm{H_{1}}$	$= \frac{\Sigma wx}{\Sigma wy}$				$w = \frac{9}{n+1}$	_		$S.E. = \left(\frac{1}{2}\right)$	$\left(\frac{pq}{wy}\right)^{1/2}$			

pean, recent Polynesian and Chilean peoples. Census at the time of the present survey numbered 1,050 natives.

Birth, marriage, and death records of the Easter Island population have been kept by the Chilean Navy for the past 50 years and by the Belgian missionaries of the Catholic church for the past 60 years. Information from these records together with information from local leaders, present missionaries, and anthropologists was utilized for classifying the subjects from whom serum was collected. Individuals any one (or more) of whose grandparents were European, recent Polynesian, or Chilcan were classified as 'admixed'; individuals all four of whose grandparents were natives of Easter Island were classified as 'pure native'. Sera were obtained from 36 'pure natives', and 123 'admixed' subjects. The total of 159 subjects constituted a random geographical sample including all the pre-existing tribes known in the last However, because of the great endogamia, the 159 individuals included many blood relatives (Table 1).

Fresh sera were prepared and stored at  $-20^{\circ}$  C until classification by vertical starch-gel electrophoresis2. Gels were stained with both benzidine and amido black 10-B.

As the sample was partially related, Cotterman's weighting gene method for calculation of gene frequency was used3; a progressively decreasing weight is given to progressively greater numbers of directly related individuals (n) examined. The weight (w) of the gene according to the size (n) of the family (1 parent and offspring) is

calculated from the formula:  $\frac{2}{n+1}$ .

In the 36 'pure natives' (Table 1) a Hp1 gene frequency of 0.86 (S.E., 0.059) was found and the distribution of observed genotypes was in Hardy-Weinberg equilibrium.

In the 123 admixed individuals (Table 1), the Hp<sup>1</sup> frequency was 0.80 (S.E., 0.028), their genotypes were also in equilibrium. This sample could be subdivided into 52 islanders with Caucasian admixture (Hp1: 0.72, S.E., 0.054), 45 islanders with Polynesian admixture (Hp1: 0.828, S.E., 0.050), 13 islanders with Chilean admixture and 13 who could not be classified with any certainty. There was a significant difference in Hp1 frequency between the 'pure natives' and the admixed individuals probably because of the small number in each category. The tendency was, however, in accordance with the expected distribution: the most divergent figures are those of the pure group and those of the islanders with Caucasian admixture.

Examination of the available data4 of Hp1 gene frequency in Pacific populations (Fig. 1) reveals two interesting phenomena: First, as Sutton<sup>5</sup> has suggested, a cline with a steady increase of Hp1 gene frequency from southeast Asia to America, passing through Alaska and progressing from north to south with a maximum value in the Araucanian Indians of Chile, is noted. Secondly, it can be suggested that another cline exists (despite needed data for Society Islands, Tumaotu Archipelago and Marquesas Islands) from east Asia through Melanesia, Micronesia and Polynesia, reaching the highest value in Easter Island. The second cline is in accordance with two others described in the same geographic distribution. The p gene frequency of the ABO blood group system also demonstrates a tendency to increase from west to east in this Shapiro's<sup>8</sup> craniological investigations suggested

that head lengths increase and head widths decrease in central Polynesia from west to east in proportion to the distance from Society Islands, and reach a maximum value also in Easter Island.

Any theory concerning this gene distribution must be speculative in view of our present ignorance of the nature and the rates of selective forces acting on Hp polymorphism. Furthermore, theories concerning the distribution in the work recorded here are complicated by migration and

possibly by genetic drift. We thank Dr. Danko Brneie and Dr. Ricardo Cruz-Coke for their advice. This work was supported by the Faculty of Medicine of the University of Chile and the Rockefeller Foundation (grant 60038), project 62-12 in a joint programme.

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### VETERINARY SCIENCE

## A Virus with Enterogenic Properties causing Degeneration of the Germinal Epithelium in Bulls

In 1959 a cytopathogenic virus was isolated in Belgium from the testes of 3 bulls showing clinical symptoms of orchitis and from 2 bulls with vesiculitis1. Since then further investigations have been made in order to assess the virological properties of the agent and the clinical picture after experimental transmission.

The virus is cytopathogenic for bovine kidney cells, porcine kidney cells, HeLa cells, and it kills chicken embryos. It has a size of about 30 mu, can be re-isolated from the fæces after an intravenous infection and is resistant to ether and chloroform.

After experimental preputial infection with tissuecultured virus in mature bulls neither febrile reactions could be detected nor other signs of illness except some ulcerations in the mouth and on the muzzle. Nevertheless, in 4 series of experimental bulls each consisting of 4 animals, the quality of the semen dropped seriously between the third week and the 7th-8th week after infection. The influence of the viral infection was especially characterized by morphological changes in the nucleus and the mid-section of the spermatozoa, by the presence of immature cells such as spermatids, spermato-