dent of the E/S ratio (Table 2). Thus it is concluded that by avoiding compression of the skin by using the techniques described, it is possible to study the properties of the skin in vitro without edge damage.

It was of interest to determine the degree to which edge damage could alter the measured electrical parameters. This was done in small chambers by determining the electrical parameters before and after compression of the same skin between the chambers. In 11 skins V_{oc} fell to 34 percent of the control value. Resistances R_1, R_2, and R_3 fell to 18, 27, and 39 percent of control values, respectively, and the I-V plots appeared to be essentially linear and showed little, if any, rectification. This observation would be consistent with the view that in the presence of edge damage to the degree produced in these studies, the measured electrical resistance would be a measure primarily of the damaged edge.

The effect of edge damage on tracer urea-^{14}C flux was also determined in seven studies and found to increase 763 percent. These data taken together show that edge damage in frog skin can be quantitatively significant and should be taken into account in the interpretation of data obtained from skins conventionally mounted with compression.

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4. Sylgard 184 (Dow Corning, Midland, Mich.), a silicone resin with added hardener, was allowed to cure in plastic petri dishes to form flat sheets 1 mm thick, from which circular gaskets with a 5-mm difference between outside and inside diameters were cut. These fit into recesses 0.5 mm deep in the faces of the chambers.
5. Bathing solution: 100 mM NaCl, 2 mM KCl, 2 mM $CaCl_2$, 1.2 mM $MgCl_2$, 0.625 mM NaH_2PO_4, 2.5 mM Na_2HPO_4, pH adjusted to 7.4 with dilute NaOH.
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Polymorphism of Soluble Glutamic-Pyruvic Transaminase: A New Genetic Marker in Man

Abstract. *Soluble glutamic-pyruvic transaminase (GPT) has three common phenotypes, each representing the homozygous and heterozygous expression of two alleles,* Gpt^1 *and* Gpt^2 *at an autosomal locus. The frequencies of these alleles vary considerably from one population to another.*

Glutamic-pyruvic transaminase (GPT), also known as alanine aminotransferase (E.C. 2.6.1.2), catalyzes the reversible conversion of L-alanine and α-ketoglutarate to L-glutamate and pyruvate. GPT resembles glutamic-oxaloacetic transaminase (GOT), malate dehydrogenase (MDH) and isocitrate dehydrogenase (ICD) in having two distinct molecular forms: one cytoplasmic (soluble) and the other mitochondrial (*1*). Genetic variation of both forms of GOT and MDH have been described in man (*2*), but there are no previous reports of such variation in either form of GPT.

Soluble GPT is particularly abundant in liver and heart (*3*); red cells also have considerable activity (*4*). The enzyme has been purified from rat liver and pig heart. The molecule, which has a mass of about 100,000 daltons, appears to consist of two subunits (*5*). Various factors, including nutritional state, pregnancy, corticoid activity, exercise, and drug administration are associated with marked changes in hepatic GPT concentration (*6*). The serum level of GPT is a very useful indicator of liver cell damage (*7*).

To demonstrate the isozymes of soluble GPT, we prepared red cell hemolyzates (*8*) and subjected them to vertical starch-gel electrophoresis at 8 volt/cm for 18 hours at 4°C in 0.1M tris-citrate buffer, pH 7.5. The GPT stain we developed was based on the chemical reactions in the spectrophotometric assay, which measures the oxidation of reduced nicotinamide adenine dinucleotide ($NADH_2$) accompanying the conversion of pyruvate to lactate by lactate dehydrogenase (*9*). Under ultraviolet light, GPT activity is detected in the gel as bands of defluorescence, representing the sites of $NADH_2$ oxidation to NAD (*10*).

Three different electrophoretic patterns were observed; they were designated GPT 1, GPT 2-1, and GPT 2 (Fig. 1). The single bands of GPT 1 and GPT 2 migrated anodally about 6 cm and 8 cm, respectively. The GPT 2-1 pattern consisted of three bands; two with the mobilities of GPT 1 and GPT 2, and a third with intermediate mobility. The GPT 2 band usually stained less intensely than the GPT 1 band.

Family studies (Fig. 2) demonstrated that the electrophoretic patterns represent the phenotypic expression of two autosomal allelic genes, Gpt^1 and Gpt^2 at the structural gene locus of GPT. The homozygous phenotypes are GPT 1 and GPT 2; the heterozygous type is GPT 2-1.

The triple band pattern of the heterozygote suggests that GPT has a dimeric structure, in agreement with previous physicochemical measurements (*5*). Thus GPT 1 and GPT 2 are dimers, each composed of two identical subunits. In the heterozygote, the slow and fast bands represent these two dimers, while the intermediate band is a hybrid dimer containing both kinds of subunits. Preliminary linkage data obtained from family studies show that the GPT locus is not closely linked with the loci of the ABO, MNS's or Rh blood groups, the haptoglobin serum group or the enzymes acid phosphatase and phosphoglucomutase-1. On the other hand, in two small families

Table 1. The GPT phenotypes and the Gpt^1 gene frequencies of blood donors from three different ethnic groups.

Ethnic groups	Number tested	GPT types			Gpt^1 frequency
		1	2-1	2	
Caucasian	253	59	133	61	0.496
Afro-American	220	146	66	8	0.814
Oriental-American	215	71	115	29	0.598

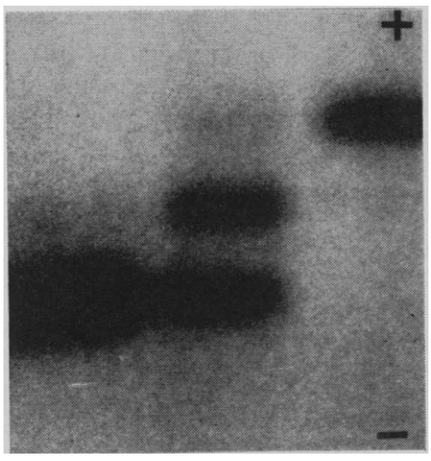

Fig. 1. Photograph of starch gel showing three electrophoretic patterns of GPT. From left to right, the phenotypes are GPT 1, GPT 2-1, and GPT 2.

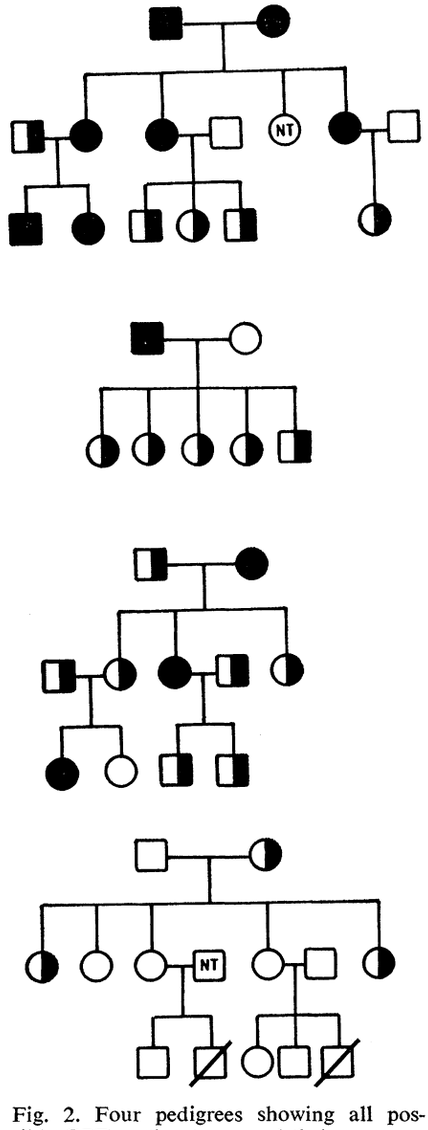

Fig. 2. Four pedigrees showing all possible GPT mating types and their progeny. The symbols are: Open, GPT 1; shaded, GPT 2; half-shaded, GPT 2-1. NT, not tested; ⊠ deceased.

with one parent heterozygous at both the soluble GOT and GPT loci, there were no recombinants out of a possible five. Further studies are required to determine whether or not these two loci are genetically linked.

Table 1 shows the incidence of GPT phenotypes among Seattle blood donors of three ethnic groups and their calculated gene frequencies. The frequency of Gpt^1 is highest in the Afro-Americans, lowest in the Caucasians, and intermediate in the Orientals, mainly of Japanese origin.

We also determined the GPT phenotypes of several hundred blood specimens obtained from natives of New Guinea, the Philippines, the Congo, Mozambique, North America (Indian), and Peru (Indian). In all populations tested, GPT was polymorphic, the frequency of Gpt^1 ranging from 0.87 in Mozambique to 0.29 in the Philippines. Thus, GPT can be added to the relatively short list of polymorphic systems with gene frequencies highly favorable to their use as genetic markers in man (*11*).

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10. The staining mixture consists of 0.1*M* tris-HCl buffer, *p*H 7.6, 200 m*M* L-alanine, 8.7 m*M* α-ketoglutarate, 1 m*M* $NADH_2$, and approximately 8 units of LDH per milliliter of solution. A filter paper (Whatman No. 1) saturated with the staining solution was laid on the bottom half of the sliced gel, which was then incubated at 37°C for 3 to 4 hours. Then the gel was photographed under a long-wave ultraviolet light with a Polaroid camera, with a yellow filter.
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Brain Serotonin Content: Physiological Dependence on Plasma Tryptophan Levels

Abstract. *Brain serotonin concentrations at 1 p.m. were significantly elevated 1 hour after rats received a dose of* L-*tryptophan (12.5 milligrams per kilogram, intraperitoneally) smaller than one-twentieth of the normal daily dietary intake. Plasma and brain tryptophan levels were elevated 10 to 60 minutes after the injection, but they never exceeded the concentrations that occur nocturnally in untreated animals as a result of their normal 24-hour rhythms. These data suggest that physiological changes in plasma tryptophan concentration influence brain serotonin levels.*

The initial step in the biosynthesis of brain serotonin involves the 5-hydroxylation of its precursor amino acid, L-tryptophan (*1*). The activity in brain fractions of tryptophan hydroxylase, the enzyme that catalyzes this reaction, is relatively low (*2*); hence, this enzyme could limit the rate at which the indoleamine is formed in vivo (*2–4*). However, the affinity of tryptophan hydroxylase for its substrate is also low (*2*), and the concentration of tryptophan usually present in neurons that produce serotonin may not be sufficient to saturate the hydroxylase (*2, 3, 5*). Thus, brain serotonin synthesis may normally be limited by the availability of tryptophan.

Experimental manipulations that markedly alter the tryptophan available to the body can raise or lower brain serotonin levels (*3*). For example, the concentration of serotonin is depressed in brains of animals given diets with