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some broad trends could be discerned from plots such as that of Fig. 2B. Other things being equal, in the 40- $\mu$  tubes the profiles tended to become increasingly more blunt as the concentration increased; they also appeared to become more blunt as flow rate and diameter decreased. These effects were much less noticeable in the tubes of larger diameter, which yielded profiles less markedly different from Newtonian profiles.

No attempt was made at this stage to assess the effect of ACD on the flow characteristics observed. Measurements made with the concentric-cylinder viscometer in the shear-rate range of 13 to 170  $\text{sec}^{-1}$  showed, however, that the ACD-treated plasma remained non-Newtonian, but the viscosity was less markedly dependent on shear rate and the values for viscosity were lower than those reported by Wells and Merrill (7) for untreated blood.

A brief comparison with results of previous investigations is pertinent.

The observation that there are two regions in the flow—a peripheral region

containing no cells and characterized by steep velocity gradients and a core in which the cells are concentrated and the velocity gradients are flatter—is in agreement with the qualitative observations of Thoma and the predictions of Coulter and Pappenheimer.

The non-Newtonian characteristics of the velocity profiles justify the concept, proposed by Haynes and Burton, of a viscosity varying across the radius. For the smaller tube sizes investigated here, however, the concept of a viscosity coefficient is to be interpreted with caution, as an indication of an average rate of momentum transfer for a homogeneous model of the flow. The presence of the peripheral layer indicates, furthermore, that the viscosity, instead of being a smoothly increasing function, varies fairly suddenly from low values near the wall to higher values in the core. Finally, occurrence of slip at the wall cannot be entirely ruled out.

The observed characteristics of the flow, such as the statistical nature of the peripheral layer and the unsteadiness of the motion of the individual

cells, are in general in close agreement with the in vivo observations of Bloch. Thus it appears that, with the proper boundary conditions, in vitro flow can indeed provide a useful model of the more complex in vivo flow (7).

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15 October 1962

## National Academy of Sciences

Abstracts of Papers Presented at the Autumn Meeting,  
29 November to 1 December, 1962, Austin, Texas.

### An Approach to the Evolution of Metabolism

Postulations that life originated by chance polymerizations of nucleotides to form macromolecules that were not only self-replicating but could also direct the formation of specific polypeptide catalysts imply that both nucleic acid and protein syntheses were antecedent to intermediary metabolism. Hypotheses more in keeping with biological concepts can be constructed from the following premises: (i) metabolic processes common to all cells living today evolved by stepwise accretion in primitive systems of "carriers" that were functional ancestors of "modern" cofactors (ADP,

NAD, TPP, THF, pyridoxal phosphate, UDP, CDP, etc.); (ii) these "proto-cofactors" which carried units of phosphate, hydrogen, active aldehyde,  $C_1$ , etc., constituted the original "bio"-catalysts and were functional before their apoenzymes evolved; (iii) the most primitive carrier-catalysts were small molecules directly derived from a few simple interrelated metabolites that constituted the innermost core of the evolving metabolic system; (iv) because the evolving pattern was determined by and dependent upon these particular carrier-catalysts derived from metabolites themselves, an autocatalytic metabolite-catalyst system developed; (v) these intermediary metabolites, besides functioning as the first

organic catalysts, also constituted the original "genetic" material because they were the agents which, when passed from primitive aggregates to daughter fragments, determined the types of reactions (hence the metabolic sequences) in the latter.

Modern RNA's are polymers of the carriers that finally evolved to transport units of energy (ADP) and the monomers used in synthesizing carbohydrates (UDP), lipids (CDP), and proteins (GDP).

A specific scheme, based on comparative biochemistry, will be presented to illustrate a postulated origin of modern metabolism from simple metabolites which could have simultaneously served as the original catalysts and hereditary materials.

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### Inhibitor of Chymotrypsin Isolated from Potatoes

This inhibitor, purified as previously described [*Proc. Natl. Acad. Sci. U.S.* (Oct. 1962)] forms with chymotrypsin, an inactive complex which has now been crystallized. When decomposed with dilute acid the crystals yield active inhibitor which has also been crys-

tallized. The crystals of the inhibitor behave as a homogeneous substance in the ultracentrifuge.

Amino-acid analyses (Moore and Stein) indicate a minimum molecular weight of about 23,000, which agrees fairly well with a preliminary value from ultracentrifuge data. However,  $\alpha$ -chymotrypsin is completely inhibited by about one-fourth its weight of inhibitor with respect to the hydrolysis of L-tyrosine ethyl ester.

The combination of enzyme and inhibitor is rapid and the complex is not appreciably dissociated. Nevertheless, when trypsin is added to the inactive complex some active chymotrypsin is formed, indicating that trypsin (in excess) displaces chymotrypsin from the complex. A large excess of chymotrypsinogen also liberates a measurable quantity of chymotrypsin from the complex, but the diisopropylphosphoryl derivatives of both trypsin and chymotrypsin are inactive.

Papain was not inhibited with respect to either esterolysis or milk-clotting. The milk-clotting activity of pepsin and that of bromelain were not affected. (Grant 8895, NIH)

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### Evidence for Stability of Magnetospheric Surface in the Solar Wind

The stability of the flow of the solar wind past the geomagnetic field is considered in the light of available experimental evidence. Magnetometer data obtained both at the earth's surface and with satellites and space probes indicates that the surface is stable. The experimental data, which demonstrates the stability, will be discussed. Some of the consequences of this new finding, as it bears on the interpretation of the planetary K index, theories of aurora, and the Van Allen radiation, will also be discussed.

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### Genetic Compatibility as Evidence of Evolutionary Relationships in North American Toads (Genus *Bufo*)

About 140 hybrid combinations involving most of the more than 30 North American species permit use of genetic compatibility data in testing hypotheses of evolutionary relationship as established from other evidence. There

is a wide variation of hybrid performance.

As arranged primarily on other evidence, the North American *Bufo* comprises 11 species groups belonging to possibly only two major evolutionary lines. All intragroup crosses have resulted in metamorphosis. In the *americanus* group (six species), 18 hybrid combinations have been made of which 14 were reciprocals of seven interspecies combinations. Of the six that have been tested (some in all possible combinations) all were fertile, although in some combinations the fertility was reduced. Similar results were obtained within the *valliceps* group (five species in area under consideration). The three species of the morphologically discrete *cognatus* group show the greatest intragroup incompatibility.

In 34 hybrid combinations, *americanus*-group females produced viable (but sterile) offspring with all males tested, including two European species, except *marinus*, which appears distantly related on other evidence. In 27 hybrid combinations involving males of the *americanus* group, the hybrids failed prior to metamorphosis except in crosses with *boreas* females, which appear closely related on other evidence.

The phylogenetic scheme postulated for the North American species is supported by hybrid performance. No reciprocal metamorphosis was found between species groups belonging to the separate evolutionary lines. All hybrids between species groups that have been tested have been sterile. Reciprocal metamorphosis of sterile hybrids occurs, however, in various interspecies-group combinations within the same line.

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### Determination of Densities and Partial Specific Volumes by Magnetic Balance Methods

Improvements in the magnetic suspension balance methods [J. W. Beams and A. M. Clarke, *Rev. Sci. Instr.* **33**, 750 (1962)] for determining densities  $\rho$  and partial specific volumes  $\bar{v}$  will be described. The values obtained for sucrose, NaCl, and KCl are in good agreement with those obtained with standard pycnometer techniques and the values using the magnetic float method reported by MacInnes and Dayhoff [*ibid.* **22**, 642 (1951); *J. Am. Chem. Soc.* **74**, 1017 (1952)]. By using a volume of 0.2 ml of the solutions and concentrations up to 1 percent, the following additional values of  $\bar{v}$  were obtained: ribonuclease,  $0.709 \pm 0.002$

ml/g; tobacco mosaic virus (TMV) in 0.01 molar solutions of EDTA, pH 7.5,  $0.738 \pm 0.002$  ml/g; and southern bean mosaic virus, 0.70 ml/g. With concentrations of TMV above 1 percent, the values of  $\bar{v}$  decrease for TMV. Curves of density versus percent concentration by weight will be given for the above substances. A single density measurement could be repeated to less than one part in  $10^4$ . The effect of concentration of EDTA in the TMV solutions also was studied. The method should be of considerable use where only small amounts of the substances are available. (Supported by USPHS grants)

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### Convergence Rates in Pseudo-Evolution

This paper derives the expected number of generations required for convergence for a certain type of pseudo-evolution. For this we assume that:

- 1) Each individual in the population is represented by a set of  $n$  binary numbers ( $n$  two-alleled "genes");
- 2) There is a population of at most two individuals at any one time which consists of an individual and its "offspring";
- 3) There is a given selection function which determines which of the two individuals has the highest survival value;
- 4) Selection is instant and absolute in that the individual with the highest survival value is kept while the other is discarded;
- 5) At each new generation a new offspring individual is born to the survivor by replication of the gene set under the influence of a mutation rate.

Bremermann ["Optimization through evolution and recombination," Proceedings on the Conference on Self-Organizing Systems held on May 22-24, 1962, at Chicago, Illinois, pp. 93-107] has shown that if the "genes" are completely independent (that is, the selection function is additive on the "genes") then the best possible single-gene mutation rate is  $\mu = 1/n$ . In this case, the time necessary to evolve to the optimal individual is, on the average,  $2.7 n \log_2 n$  generations.

In this talk we define the concept of  $s$ -lethal dependency and show that the best mutation rate is  $\mu = k_s/n$ , and the expected number of iterations required for convergence is approximately  $C_s n^s$ , where  $k_s$  and  $C_s$  are constants depending on  $s$  only.

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## Structural Differentiation in Chromosomes of *Gossypium*

Chromosomes of *Gossypium* species fall into five genomes in terms of metaphase pairing and chromosome size. In intergenomic hybrids metaphase bivalents are reduced in number and usually one-chiasma, but at pachytene chromosomes are completely paired and equal in length, as in species. These observations led to a study of intervening stages of meiosis in species and hybrids.

At diplotene and diakinesis chromatic and achromatic regions as well defined. Each genome, and each chromosome within a genome, has a characteristic amount and distribution of chromatic material. At diplotene in intergenomic hybrids, homologous chromosomes are paired but differ in chromatic sections. As meiosis proceeds, chromatic and achromatic regions contract differentially, and paired chromosomes without chiasmata fall apart. In intergenomic hybrids, chromosomes of bivalents which remain at diakinesis and metaphase are markedly unequal. Achromatic regions change from a long fibrillar structure to short diffuse masses, and at metaphase form a negligible portion of the chromosomes. Chromatic regions become compact and form the bulk of the metaphase chromosome. Size and shape of metaphase chromosomes are thus determined largely by amount and distribution of chromatic material. Chiasmata occur most frequently in achromatic regions. Therefore neither chiasma formation nor breakage frequency need be proportional to metaphase chromosome length. Genome differences between large (A genome) and small (D genome) chromosomes of *Gossypium hirsutum* nevertheless occur in the recovery and transmission rates of chromosome aberrations, and in number and position of chiasmata. Study of monosomics, translocations, haploids and asynaptic stocks confirms the above observations and conclusions.

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## DNA-Histone Association as a Basis for Alternative Hereditary Gene States

Theoretical considerations demand, and experimental analyses confirm that histones constitute a more homogeneous group of substances than do the DNA's with which they associate. Thus within one cell, different stretches of DNA may ultimately be grouped on the basis of common histone associations. During the life cycle of organisms, cells are

found in which the main complement of histones has been replaced; this indicates that variation in histone association may occur among homologous genes at different times during development, thus within different cells, as well as among different groups of genes within the same cell. If histones synthesis is controlled by specific genes, the DNA-histone association provides a means whereby histone-producing genes may variably influence histone associating genes.

DNA replication must occur amidst the spectrum of histones found within the cell. Unless the DNA-histone association is random, the newly replicated DNA would be expected to associate with the same histone after replication as before replication. It is unlikely that the associations are restricted by inherent differences in affinities of DNA for histone. The factor which determines the specificity of complexing may ultimately lie with the association which exists prior to replication. The complex should then replicate as a unit, the DNA would act as its own template, and the preexisting histone would determine the association of the newly replicated DNA with the proper previously synthesized histone.

In this manner, the DNA-histone association may provide a basis for alternative hereditary states of the gene, known to exist, which are moderately stable and potentially reversible.

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## Level Structure of the Mirror Nuclei $\text{Ca}^{41}\text{-Sc}^{41}$

Recent measurements of the  $\text{Ca}^{40}(\text{p,p})\text{Ca}^{40}$ ,  $\text{Ca}^{40}(\text{d,p})\text{Ca}^{41}$ , and the  $\text{Ca}^{40}(\text{p},\gamma)\text{Sc}^{41}$  reactions together with results from earlier experiments have led to a fairly complete picture of the level structure of the mirror nuclei  $\text{Ca}^{41}\text{-Sc}^{41}$  up to an excitation energy of about 7.5 Mev. Approximately 100 states identified in this interval have been assigned widths ( $\Gamma$ ) and  $I(J)\pi$  values. Such information is of interest in connection with the location and amount of fragmentation of the single particle states of  $2p_{3/2}$ ,  $2p_{1/2}$ ,  $2d_{5/2}$ ,  $1g_{9/2}$ ,  $1f_{5/2}$ , and  $3s_{1/2}$  character which are expected in or near this energy range. The principal features which emerge from an analysis of the available data are: (i) The principal part of the  $p_{3/2}$  and  $p_{1/2}$  single particle strength is concentrated in single states occurring below 4 Mev. (ii) In the range of excitation energy below 4 Mev for which the data on both nuclei are most complete, the correspond-

ence in excitation energies is generally within 80 kev. A striking exception to this pattern is presented by the principal  $p_{3/2}$  and  $p_{1/2}$  states which occur in  $\text{Sc}^{41}$  220 kev and 490 kev lower in energy respectively than in  $\text{Ca}^{41}$ . (iii) The d-states appear in significant numbers above 4 Mev, and f-states above 5 Mev. The s-states appear infrequently throughout the energy range. Weak evidence for g-states is found between 5 and 6 Mev. (iv) The individual reduced widths measured are of the order of 1 percent or less of the Wigner limit. The summed d-, f-, and s-state strengths are respectively less than 15 percent, 20 percent, and 3 percent of the Wigner limit which places the "center of gravity" of these states at excitation energies beyond the range of the present data.

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## Ultrasonic Measurements in Superconductors

In pure metals there is an appreciable attenuation of high frequency ( $\geq 10$  mcps) sound waves at low temperatures due to the interaction of the lattice and the conduction electrons. This interaction has led to the use of ultrasonics in studying electronic properties. It can be shown that the magnitude of the electronic attenuation increases with frequency and electron mean free path and depends strongly on such parameters as effective mass, Fermi velocity, and the electron lattice interaction constant. In normal metals, the use of ultrasonic attenuation in the presence of a magnetic field has been used to map the Fermi surface in many single crystalline materials.

Ultrasonic attenuation studies in "soft" superconductors have proved to be equally fruitful. As the temperature is lowered below the transition, the electronic attenuation of both compressional and shear waves in soft superconductors decreases from its value at the transition temperature, reflecting the decrease in the normal electron density. The shape of the attenuation versus temperature curve at low temperatures can be used to measure the energy gap predicted by the Bardeen-Cooper-Schrieffer theory. The transition is usually quite sharp and provides a tool for making accurate measurements of critical fields and currents as a function of temperature.

The authors have recently made a study of the temperature variation of the attenuation in niobium and ni-

biium-zirconium alloys which have "hard" superconducting properties. In the moderately pure niobium the attenuation was found to increase as the temperature was lowered. The temperature at which this increase begins is frequency dependent. In the case of the niobium-zirconium alloys, irregular variations of the attenuation with temperature were observed. It was found that these variations were approximately periodic when the attenuation was plotted versus the inverse of the critical field. The period of these oscillations is found to scale with the wavelength of the sound wave. Further investigations of this periodicity as a function of frequency and alloy concentration are currently being conducted. A theoretical model has been proposed by the authors to explain this behavior.

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### Unequal Crossing Over in Salmonella

The mutant *argA-162* was found to be a "selfer," that is, it gave rise to wild-type (+) variants in self-transduction with homologous phage. Tests showed that the following explanations cannot account for the + variants: contaminated phage, presence of *argA+* transducing fragments in the phage, suppressor mutations, an unusual association between the transducing fragment and the bacterial chromosome, suppression of growth of revertants on the control plates, or greater residual growth of bacteria on the experimental plates. It was found that factors which increase the efficiency of transduction (number of bacteria, phage multiplicity, and ultraviolet irradiation of phage) also increase the number of + variants. It is proposed that unequal crossing over, involving adjacent bases of the DNA of a gene, is responsible for this phenomenon.

Tests of 166 mutants, representing 13 gene loci, identified 55 as selfers. The mutants tested included 49 induced by 2-aminopurine, of which 51 percent proved to be selfers. Of the remaining 117 (some spontaneous, others induced by ultraviolet, x-rays, neutrons, nitrous acid, or diethyl sulfate), 27 percent are selfers. Therefore the occurrence of selfers among *Salmonella* auxotrophs is not a rare event; and it seems likely that the A nucleotide is involved. (Under auspices of the AEC)

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### Terrestrial Abundance of Thorium and Its Availability as a Nuclear Fuel

The availability of thorium, particularly relative to uranium, is important in evaluating various systems for producing nuclear-fission energy. Analysis of rocks and meteorites and the distribution of radiogenic leads in geologic time indicate a crustal abundance of thorium four times that of uranium (12 and 3 parts per million, respectively). Despite its greater abundance, the availability of thorium, as measured by ore reserve estimates, has not increased in the last decade as markedly as that of uranium. Prior to the present work, it was often concluded that the greater availability of uranium was due to the greater effectiveness of natural mechanisms in concentrating uranium; an alternative hypothesis would be to attribute the greater availability to greater exploration effort on uranium.

In the last 2 years over 10<sup>7</sup> tons of recoverable (less than \$100 per pound) thorium have been indicated in the Conway granite of New Hampshire. The primary emplacement of thorium, the development of advanced radiometric equipment to determine thorium in the field and in the laboratory, and appropriate statistical techniques, all contributed to the quick and cheap development of these reserves. Given equal exploration effort in all regions, these results suggest that the availability of thorium may approach or even exceed that of uranium.

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### Retention Time Relationships in the Gas Chromatographic Separation of Steroids

"Steroid numbers" have been defined (W. J. A. VandenHeuvel and E. C. Horning, *Biochim. Biophys. Acta*, in press) in terms of relative retention time relationships determined for a given steroid and two reference compounds (androstane and cholestane). Hypotheses developed earlier for the paper partition chromatographic separation of steroids, when applied to gas-liquid chromatography, lead to the expression:

$$SN = S + F_1 + \dots + F_n$$

where SN is the steroid number, S is the carbon content of the steroid skeleton, and  $F_1 \dots F_n$  are values characteristic of the functional groups of the steroid. While steroid numbers are not different in principle from relative retention times, they are far more useful in experimental work. These values are not dependent upon the

temperature of the separation over the range 195°–215°C (steroid numbers to about 29).

This work has been extended in several ways. Through the use of appropriate reference compounds steroid numbers up to about 35 have been determined (temperatures to 235°C). The variation in steroid number values, determined with different reference compounds and over the temperature range 195°–235°C, is about 0.1. The possibility of defining temperature-independent relationships for selective phases has also been investigated. Effects due to positional and stereochemical arrangements of functional groups may be studied by these methods, by the T-value method of Haahti, VandenHeuvel, and Horning [*Anal. Biochem.* **2**, 344 (1961)] and through the  $\Delta R_M$  values of Knights and Thomas [*Nature* **194**, 833 (1962); *Anal. Chem.* **34**, 1046 (1962)]. The possibility of extending steroid number procedures to compounds other than steroids has been explored for a few examples.

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### Cosmic Ray Muons at High Energies

Absolute directional intensities of cosmic ray muons have been measured with an extensive telescope for charged particles. The telescope consists of about 100 large Geiger counters arranged in crossed configurations to determine the directions of very penetrating particles incident on the apparatus. The particles detected are known to be muons from the decay of extremely energetic cosmic ray secondaries very high in the earth's atmosphere. Most of the measurements were made below hundreds of feet of limestone of well-known chemical composition. Minimum muon energies required to reach the apparatus from the surface ranged from about 10 to 100 billion electron-volts. It is found that directional intensities of muons with energies above about 40 billion electron-volts are greater from near horizontal directions than from the vertical. This and other aspects of the cascade processes which give rise to the energetic muons in the atmosphere are interpreted theoretically by means of a diffusion equation which describes the interaction and decay of muon parent particles. (Assisted by J. A. Smith, W. R. Sheldon, O. L. Smith, R. R. Kasten)

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## An Extension of the Sampling Theorem

Regular (cyclic) sampling of a time-dependent function cannot reduce aliasing. It yields information which differs from that obtained by equispaced sampling, but it does not yield more information. An elementary demonstration based on a Taylor expansion of the function is given.

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## Induction of Motility in the Spores of Some Actinoplanaceae (Actinomycetales)

The Actinoplanaceae, recently discovered family of the Actinomycetes, is based at present on two genera, *Actinoplanes* and *Streptosporangium*. In the former the spores are motile while in the latter motile spores have never been observed. We have in manuscript descriptions of two new genera, one with rod-shaped spores which are usually rather poor swimmers in water, the other with spiral spores in which motility in water mounts is rare or at best languid.

To carry out studies on the structure and behavior of the motile cells it was essential that we have them in large numbers. We accidentally discovered that 1 percent casamino acids would induce motility in the spiral and rod-shaped spores. We also found that L-arginine hydrochloride (6 mM), as used by certain bacteriologists, and urea (0.01 M) greatly increased motility.

Electron micrographs showed a single polar flagellum on the rod-shaped spores. Flagella were also shown on the spiral spores but their number and position of attachment is uncertain.

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## Growth-Inhibition Studies Employing Peptides and Their Analogs

$\beta$ -2-Thienylalanine (I), a structural analog of phenylalanine (II), and 13 synthetic peptides containing I are effective inhibitors of microbial growth. The corresponding peptides of II, including di-, tri-, and tetrapeptide structures, can replace II in stimulating microbial growth, and are more effective than II in promoting growth which has been suppressed by addition of I to the medium. Peptides of I are more toxic than I when tested in a system where growth is dependent

upon a supply of exogenous peptides of II.

Specificity in utilization of di-, tri-, and tetrapeptides by *Escherichia coli* strain 9723 can be demonstrated in a system where growth is inhibited by adding to the salts-glucose medium 3000  $\mu$ g of I and 70  $\mu$ g of II per 5 ml. (Addition of another 100  $\mu$ g of II is required to stimulate growth in the presence of 3000  $\mu$ g of I). Growth which is stimulated by addition of 10 units (one unit contains 1  $\mu$ g of phenylalanine or  $\beta$ -2-thienylalanine) of glycylphenylalanine per 5 ml is inhibited by 1000 units of glycyl- $\beta$ -2-thienylalanine (III), but not by a similar amount of I, diglycyl- $\beta$ -2-thienylalanine (IV) or triglycyl- $\beta$ -2-thienylalanine (V). Growth stimulated by diglycylphenylalanine is inhibited by IV, but not by I, III, or V. Growth stimulated by triglycylphenylalanine is inhibited by either IV or V, but not by I or III. Possible metabolic implications will be noted.

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## Effect of Invertebrate Hormones on Vertebrate Tissues

After noting an increased incidence of hereditary melanotic tumors in *Drosophila* when hormones of the ring gland were diminished, the effects of brain hormone, farnesol, and ecdysone on mammalian tissues were tested by injections in vivo, observations on cells in tissue culture, and by measuring the rate of protein synthesis. Farnesol administered in doses of 0.1 to 0.2 ml, in from one to five doses in mice bearing sarcoma 180, resulted in 25 mice with complete regression, 21 mice with partial regression, and 39 mice with no impairment of growth of the tumor. Early death occurred in 35 additional animals. Three complete and two partial regressions occurred in the control group of 53 animals. Five complete and 1 partial regression of transplanted tumor occurred after injection of 400 to 2400 *Calliphora* units of ecdysone in comparison to none in the control group. Only six mice with transplanted sarcoma 180 were treated with brain hormone. Results were identical with the control group. When L cells were grown in medium containing various concentrations of ecdysone, increasing concentrations reduced the numbers of cells counted. In amounts of 15,000 c.u. per ml or greater, all cells were dead within 48 hours. No effect was noted with 100 c.u. per ml. In order to test whether ecdysone has more than a nonspecific toxic effect on mammalian tissues, the fraction of human and

murine liver obtained by centrifugation at 14,000g was incubated with DL-leucine-1- $C^{14}$  and the rate of incorporation into protein determined by counting in the proportional range. There was an increased rate up to 92 percent with threshold between 100 and 250 c.u. and increased effect with increased dosage. In view of the action of ecdysone on initiating the formation of puffs at specific sites on the salivary chromosomes in insects, this action in augmenting the rate of protein synthesis in mammals is of particular interest in relation to differentiation and the control of growth.

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## Hydromagnetic Stability at a Fluid Velocity-Discontinuity

The stability of a sheet that separates two perfectly conducting, inviscid, compressible fluids, and across which the tangential component of the fluid velocity and the magnetic field change discontinuously, is examined.

It is shown that the sheet is unstable in the absence of a magnetic field. The formal conditions for stability in the presence of a magnetic field are derived and are investigated in detail for certain special cases of interest.

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## Eigenprojector Theorem

The eigenprojector theorem [see F. A. Matsen, *Finite Group Algebras and Quantum Mechanics*, Tech. Rept., Molec. Physics Group (Univ. of Texas, Oct. 1962)] states that if a Hamiltonian commutes with algebra  $A$  of finite group  $G$ , its eigenfunctions are given by

$$\psi^j_{zt} = \mathbf{Z}^j_t \Phi_z \quad t = 1 \text{ to } a^j$$

where  $\Phi_z$  is an asymmetric (primitive) function and  $\mathbf{Z}^j_t$  is an eigenprojector which is (i) an idempotent element of the integral (simple matrix) subalgebra  $A^j$  of  $A$ , (ii) an element that transforms under left multiplication by elements of  $A$  according to the  $a^j$ -dimensional irreducible representation  $\Gamma^j$  of  $A$ , (iii) an eigenelement to the class operators with eigenvalues which define the  $j$ th symmetry state. The Hamiltonian matrix elements are independent of  $t$ , so the  $j$ th symmetry state is  $a^j$ -fold degenerate. The eigenprojector is a linear combination of



$a^j$  independent basis elements with coefficients determined, not by the symmetry, but by the detailed interactions in the Hamiltonian. The eigenprojector theorem places the finite symmetry problem of quantum mechanics into the group algebra.

For an  $N$ -fermion system, the spin-free Hamiltonian is invariant under the group of permutations of the space coordinates of the fermions. The space permutation states and the space eigenprojectors are characterized by partitions of  $N$ . Expectation values for spin-free operators are formulated in terms of the space eigenprojectors without explicit use of spin. The permutation algebra and the space eigenprojector have a structure basis, each element of which is characterized by a structure composed of integers and tie lines. In those structures which are admissible by the Pauli principle for polyelectronic systems, no more than pairs of integers may be tied. Each tie line corresponds to an electron pair, each basis element of the structure basis corresponds to a chemical structure and the eigenprojector itself corresponds to a superposition or resonance hybrid of chemical structures. The formulation makes explicit the relationship between chemical concepts and the abstract permutation algebra.

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#### **Possible Trisomy in Chromosome Group 6 to 12 in a Mentally Retarded Patient**

Except for the X-chromosome, occurrence of a supernumerary chromosome in the group of submetacentric chromosomes made up of the X and pairs 6 to 12 has been seldom described. Trisomy is better known among the shorter chromosomes, as in the 21-trisomy of the Down syndrome. A possible trisomy in group 6 to 12 has now been found in a mentally retarded patient of the Austin State School.

Peripheral leukocytes brought to division *in vitro* by means of phytohemagglutinin made up two modal groups with respect to chromosome number. The patient is therefore to be considered a chromosomal mosaic. Slightly less than half the dividing leukocytes had the normal number of 46 chromosomes, with no apparent abnormality in the individual elements. The other modal group of leukocytes contained 47 chromosomes each, the extra one being a submetacentric chromosome slightly shorter than others in the group 6 to 12. For this reason, and because neutrophil leuko-

cytes in smears did not exhibit the "drumsticks" nor buccal smears the sex chromatin typical of females, the extra chromosome was not thought to be an X-chromosome.

Besides mental deficiency, the patient, a 54-year-old negro male of short stature, was marked by a pronounced pot-belly, depressed bridge of nose, curvature of the terminal phalanx of the little finger, acromicria, hypogenitalia, undescended testicles, scoliosis, a mild kyphosis, rather thick skull bones, with the "tower skull" deformity, mild diabetes, and osteoarthritic changes around the knee joint.

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#### **Significant Liquid Structures**

The important problem of liquid theory is the development of a believable model of the liquid state which leads to partition functions for the normal and activated states which quantitatively account for thermodynamic and transport properties. Although a number of models have been formulated which account reasonably well for the behavior of fluids in the imperfect gas range and for critical properties they are not as a rule simultaneously adequate for melting properties. The most nearly adequate model covering the entire fluid range has been called "Significant Structures."

Since the Helmholtz Free Energy,  $A$ , can be expressed as

$$A = -kT \ln \int \frac{e^{-H/kT} dq_1 \dots dp_{2N}}{h^{2N} N!}$$

where the Hamiltonian,  $H$ , is the known sum of the kinetic and potential energy for the system, and since when  $A$  is known all thermodynamic properties are calculable, a model may seem superfluous. It would be superfluous if one could carry out the integration. A model greatly simplifies the integration by restricting the part of phase space which must be integrated over.

Melting occurs by the fluidization of vacancies in a modified solid structure. Isolated vacancies do not provide enough entropy to give  $T\Delta S = \Delta H$  where  $\Delta H$ ,  $\Delta S$ , and  $T$  are the enthalpy, entropy, and temperature of melting, respectively. However, at an above the melting point as enough vacancies are added there is a loosening of the liquid structure until the vacancies approximate in freedom of motion molecules in the gas phase. As a result vacancies in the liquid approximately

mirror the behavior of molecules in the vapor phase. The quantitative consequences of this model, first outlined by the author in 1936, will be traced as time permits.

HENRY EYRING

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#### **Zero Point of Charge of Solid Metals in Contact with Electrolyte Solution**

Until recently, probing the electrical double layer between metal and solution was unrewarding except with liquid metals. Now, with the availability of metals in single crystal form and by the use of pulse techniques, information on the electrical double layer of a more meaningful kind has become available. From these measurements reasonable, although still debatable, values of zero points of charge have begun to emerge. This quality, which is certainly involved in the kinetics of electrode processes, is now seen quite clearly to be a property not related simply to the work function of the metal but, instead, to the whole system. This suggests that specific adsorption is a major factor in determining the variation of potential with distance between metal and solution.

NORMAN HACKERMAN

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#### **Therapeutic Effect of Bone Marrow Transplants Following Lethal Irradiation**

Chicks 17 days of age, when exposed to 1800 r of discontinuous x-irradiation (40-minute interval between each 300 r) suffered their initial mortality after 5 days postirradiation, with approximately 100 percent mortality occurring by 14 days postirradiation.

Intravenous injections of bone marrow homogenate obtained from randomly selected nonirradiated birds significantly reduced 30-day postirradiation mortality. Variations in the cell concentration of bone marrow homogenate injected also influenced survival with 2-, 4-, 8-, and 16-percent suspensions resulting in 30-day postirradiation mortality values of 55, 19, 12, and 6 percent, respectively, as compared with 86 percent mortality for irradiated noninjected controls.

Blood hematocrit values for irradiated controls and for irradiated groups receiving bone marrow homogenate began to decrease by the third day postirradiation and reached a low value (10) after 2 weeks, with surviving birds returning to normal (29) by

30 days postirradiation. This decreased red cell volume was accompanied by an increase in plasma volume to the extent that total blood volume remained essentially the same as that of the nonirradiated controls.

Selection of donor and irradiated host birds on the basis of blood group genotype showed incompatibility of five blood group systems, and resulted in mortality essentially the same as that of the irradiated noninjected controls (79 percent). This compares with 32-percent mortality for birds injected with marrow of compatible blood types. Mortality was further reduced to 16 percent when donor and irradiated host birds were full sibs in addition to having compatible blood group genotypes.

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#### **Inhibition of Protein Synthesis and Some Quantifications of Early Estrogen Action and Response**

Mueller *et al.* (1961) have reported in vivo studies that combined treatments of individual, ovariectomized rats with puromycin (a known inhibitor of protein synthesis) and 10  $\mu\text{g}$  of estradiol-17 $\beta$  lead in 4 hours to the following situations for previously atrophic uteri: (i) suppression of protein synthesis to levels below control levels as well as below those expected to result from estrogen stimulation alone and (ii) retardation of the increases in RNA and phospholipid synthesis which normally follow such hormonal stimulation. Suppressions of the latter two below control levels were not observed. That these workers injected doses of estradiol far above those expected to be operative at the tissue level (10  $\mu\text{g}$  being equivalent to an amount sufficient for induction of vaginal estrous in over 100 ovariectomized rats) raised the possibility that their findings reflect a nonphysiological situation. Their work has been repeated utilizing lower doses of estradiol, and, in addition, of two other naturally occurring estrogens, estriol and estrone. Minimum doses required for in vivo initiation of the estrogen response by these steroids were measured by determinations of the incorporations of glycine-2-C<sup>14</sup> into uterine protein and of orthophosphate-P<sup>32</sup> into uterine RNA.

For test periods of 4 hours, minimum doses necessary for acceleration of protein synthesis above that of controls are as follows: estradiol, 0.03

$\mu\text{g}$ ; estriol, 0.05  $\mu\text{g}$ ; and estrone, 0.6  $\mu\text{g}$ . In each case, acceleration of RNA synthesis was observed. For the range of hormone doses tested, no evidence was found for estrogen-accelerated protein synthesis without acceleration of RNA synthesis. Using the hormone doses noted above, combined treatments of estrogen and puromycin resulted in the same picture for the early estrogen response in the uterus as found by the workers cited. The results are compared with the findings of Hisaw (1959), who estimated minimum doses necessary for estrogen stimulation of the rat uterus by determination of the minimum doses required for uterine imbibition of water (a process now known to be blocked by puromycin). Although there are some quantitative differences, the data reported complement qualitatively Hisaw's findings.

Since the findings here reported confirm at a lower level of hormonal stimulation the studies of Mueller and his co-workers, such are considered to support strongly their thesis that protein synthesis is mandatory for the early estrogen action in the uterus, and the view is favored that selective control of enzymatic protein synthesis is a key aspect of the inceptive action of estrogen on the metabolic apparatus of uterine tissue. Progress in this area is considered to depend upon the discovery of a cell-free, estrogen-sensitive, protein-synthesizing system.

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#### **Longitudinal Differentiation of Chromosomes and the Possibility of Interstitial Telomeres**

It has been found previously that the thymidine analog, 5-bromodeoxyuridine (BUDR) damages mammalian chromosomes at specific regions. One of the regions that respond to BUDR preferentially is the telomere. In the Chinese hamster, it is found that chromosome No. 1 contains an interstitial region (region 7) which is extremely susceptible to BUDR toxicity. These regions are interpreted as areas rich in adenine and thymidine (A-T) pairs.

When the Chinese hamster cells are x-irradiated (250 rad), the distribution of damage along chromosome No. 1 is found to be at random if the samples are taken 4 hours postirradiation (G<sub>2</sub> cells). However, if samples are taken 8 or 12 hours postirradiation (S or G<sub>1</sub> cells respectively), the distribution is not at random, with region 7 showing a distinct peak (approximately 25 percent). The nonrandom distribution of breaks is thus

regarded as a result of nonrandom restitution, not nonrandom induction. Since region 7 is considered A-T rich, a break in this region would retain sufficient A-T pairs to serve as a new telomere. Thus interstitial A-T regions may be regarded as interstitial telomeres.

To test this hypothesis further, it is anticipated that region 7 of chromosome No. 1, after induced breakage by BUDR, does not require rejoining. Thus in the ensuing cell generations, one of the two chromosomes No. 1 will be replaced by a new submetacentric element whose long arm should be equivalent to the short arm of chromosome No. 1 and the short arm equivalent to the length between region 7 and the centromere. This was found to be the case. The phenomenon supports the hypothesis that region 7 is an interstitial telomere.

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#### **Papovaviruses, a New Grouping of Tumorigenic Agents**

Recent studies show that four tumorigenic viruses fall into a natural group, whose name is derived from the first two letters of the name of each member virus: papilloma (of rabbits and of man), polyoma (of mice), and vacuolating (SV-40 of monkeys). The cardinal features of the papovaviruses are: similarities in size (40–50 m $\mu$ ), morphology (42 capsomeres), and buoyant density in CsCl (1.30); presence of double-stranded DNA; absence of essential lipid; relative thermal stability; enhanced inactivation by divalent cations; slow growth cycle (about 24 hours) characterized by multiplication within the nucleus and involvement of the nucleolus. Other viruses with similar properties except for tumorigenic activity have been added to the group.

The simian papovavirus has been injected or fed inadvertently to hundreds of thousands, if not millions, of persons as a contaminant of polio and adenovirus vaccines made in monkey kidney cultures. Although low-grade infections have occurred in man, no illness has been observed. In newborn hamsters, fibrosarcomas are readily produced. An important property of the virus is its ability to transform normal human fibroblasts in vitro to epithelioid cells characterized by an abnormal growth pattern, accelerated growth, and chromosomal alterations [H. M. Schein and J. F. Enders, *Proc. Natl. Acad. Sci. U.S.A.* **48**, 1164 (1962)].



In monkeys, the natural host of the virus, a latent infection was induced [A. Ashkenazi and J. L. Melnick, *Proc. Soc. Exptl. Biol. Med.*, in press]. This was characterized by virus in urine, followed by antibodies in the blood and in the urine, and chronic localization of virus in the kidneys. Virus could not be recovered from kidney cells by rupturing them, but only by growing biopsies in tissue culture. After 3 to 4 weeks of culturing, such biopsies spontaneously developed the vacuolating type of degeneration characteristic of SV-40, and the virus was then readily detected by passage and typing. The same monkey yielded the virus from repeat kidney biopsies over a period of 8 months.

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### Allegheny Paleogeography in the Northern Appalachian Plateau

The middle Pennsylvanian Allegheny formation, a 300-foot-thick rock unit outcropping in the northern part of the Appalachian Plateau, records some of the early geological events which culminated in the late Paleozoic Appalachian orogeny. This record is essentially depositional and consists of sedimentary rocks representing swamp, fluvial, and lacustrine systems, and marine and brackish water embayments developed on a shifting coastal plain. Large-scale shoreline migrations seem to be determined, at least in part, by gross overall patterns of differential subsidence, quantity of sediment locally available, and character of the previously deposited sedimentary substratum. The overall pattern of deposition seems to suggest sources of sedimentary supply from the north, east, and south. The general environmental pattern is one of gradual northward and westward withdrawal of marine embayments from older to younger Allegheny time.

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### Compositional Aspects of the Nucleic Acids of Cotton

The nucleotide composition of isolated DNA and fractional and total RNA from wild and cultivated species of the genus *Gossypium* was studied.

All of the DNA's were of the adenine-thymine type, being higher in these bases and lower in cytosine plus

5-methylcytosine and guanine contents than those of other reported plant DNA's. In general the overall DNA compositions of the 13 species examined (four cultivated, nine wild) exhibited little or no species specificity. In contrast the cytosine/5-methylcytosine ratios alone, in the wild species, plus two of the cultivated species (all diploids), tended to be species specific. The two other cultivated species (tetraploids) were not similarly differentiated. Compositional data on the DNA's of doubled polyhaploids of two varieties of *hirsutum* showed that their cytosine/5-methylcytosine ratios, while not differing from each other, were significantly higher than those of the ordinary genetic types.

Total RNA, of which no satisfactory isolates were obtained, was determined without prior separation from the tissue. It was of the guanine-cytosine type, as are all of the plant RNA's investigated, and its composition was fairly uniform throughout the 13 species.

Two RNA fractions, differing in adenylic and cytidylic acid contents, were isolated in purified form and their compositions in the four species studied were different.

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### Microwave Spectrum of $O^{18}$ -Nitrogen Dioxide

Ten hyperfine components of the  $8_{08} \rightarrow 7_{17}$  and nine hyperfine components of the  $9_{10} \rightarrow 10_{0,10}$  rotational transitions of  $N^{14}O^{16}O^{18}$  have been measured. By fitting the microwave spectrum of  $N^{14}O_2^{16}$ , and using the same Fermi, magnetic dipole, and quadrupole coupling constants for  $N^{14}O^{16}O^{18}$ , the hyperfine structure of  $N^{14}O^{16}O^{18}$  is fit within experimental error. The spacings between triplets for the  $23_{2,22} \rightarrow 24_{1,23}$  and  $40_{3,38} \rightarrow 39_{3,37}$  transitions of  $N^{14}O_2^{16}$  are in error by about 15 Mc. Otherwise the normal species is fit within experimental error. The difficulty with the triplet spacings is perhaps ascribable to approximating the second order perturbation term in the spin rotation interaction by that obtainable from Wang wave functions.

The Fermi constant of  $146.64 \pm .8$  Mc obtained here is very close to that determined by Baird and Bird and corresponds to about 10 percent 2sN for the odd electron. The magnetic dipole interaction constants indicate that the odd electron has about 32 percent of

2pN with the p orbital directed along the symmetry axis. The spin-rotation interaction constants show about the same isotope dependence as the rotational constants, as is expected. At present the estimated error in nuclear quadrupole coupling constants is about the magnitude of the constants.

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### Uridinediphosphogalactose 4-Epimerase Inhibition in Mammalian Cell Cultures as Correlated with Aerobic Glycolysis

Previous studies have demonstrated that mouse ascites tumor and Earle's established L-cell cultures from mouse connective tissue are unable to use galactose as a hexose source (Eagle, Racker). It has been suggested that UDPGal 4-epimerase activity is lacking in L-cell cultures as well as in extracts from such cells (Maio, Rickenberg). We have found that extracts from L-cell cultures contain 4-epimerase if tested under optimal pH and with optimal concentrations of DPN. However, L-cell epimerase as well as purified calf liver epimerase can be strongly inhibited under conditions which may well exist inside cells, especially under conditions of aerobic glycolysis. It turns out that the previously described inhibition of mammalian epimerase by DPNH (Maxwell and Kalckar) is strikingly intensified at a pH of 7.5 or a more acid pH. Under these conditions a mere trace of DPNH in the presence of excess DPN strongly inhibits the epimerase. Studies on intact HeLa cells under conditions of aerobic glycolysis strongly indicate that the epimerase step in these cells is likewise blocked. These observations may indicate a relation between the Crabtree effect and the metabolic block of epimerase in tumor cells.

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### An Analysis of Mutations Confined to a Small Region of the X-Chromosome of *Drosophila melanogaster*

In *Drosophila melanogaster*, the very closely linked and functionally related genes, white (*w*) and zeste (*z*), can also be shown to exhibit synaptic affinities for one another. The region between the two loci repre-

sents a genetic length about 0.5 percent crossing over and contains about 12 bands on Bridges salivary-gland-chromosome map. This region was singularly devoid of known mutant sites until this investigation was undertaken.

Since it is possible to look upon zeste and white as components of a single functional unit, it is of considerable interest to examine the nature of mutations which occur in the region bordered by these components.

The employment of an efficient screening technique has yielded 22 separate mutations in this strictly limited region of the chromosome. All of them act as recessive lethals.

The genetic analysis of 16 of the mutants shows they are distributed among five distinct functional units (complementation units) in the chromosome. These five units do not include the original white and zeste loci, and they represent a minimum number that may be discovered as saturation of the region is approached.

Detailed analysis of the function of the mutants of this region along with their spatial distribution in the chromosome will give valuable information concerning the relationship between organization and function in this chromosome region. Thus far there is no indication that any of the five groups are functionally related to either zeste or white despite their very close position in the chromosome.

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## Quantitative Aspects of

### Virus Inhibition by Interferon

Interferon is a protein capable of provoking in mammalian cells an intracellular resistance to viral infection. Using an interferon prepared from the PR8 strain of influenza virus, chick embryo tissues, and western equine encephalomyelitis virus, the following concepts will be discussed: (i) the nature of the resistance to virus infection; (ii) some considerations of concentration of interferon and time on the ensuing resistance; (iii) the preliminary localization of the metabolic pathway blocked by interferon action. It will be shown that both inhibition of virus production and protection against cytopathic effects can result from treatment of cells with interferon. Inhibition of virus production is a more sensitive measurement of interferon action. The time necessary to reach a given level of virus inhibition or to reach protection against cytopathic effects is a function

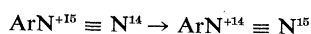
of the concentration of interferon suspension added to cultures and the time permitted for its uptake. The duration of interference in interferon-treated cells is also a function of the dose of interferon received by cultures. By manipulating the dose and time of interferon addition to virus-infected cells, evidence was obtained indicating that interferon inhibited virus production by blocking new viral ribonucleic acid synthesis.

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### Tracer Study of Diazonium Salt Decomposition

Benzenediazonium- $\alpha$ - $N^{15}$  fluoborate (about 95 percent  $N^{15}$ ) was prepared by way of benzamide- $N^{15}$ , and aniline- $N^{15}$ , followed by diazotization with ordinary nitrous acid. A degradation scheme was devised based on the reaction of the diazonium ion with azide ion, the course of which is known. This degradation gives two different nitrogen gas samples from which two independent values of the  $N^{15}$  content of the  $\beta$  position can be obtained. The labeled diazonium salt subjected to the degradation showed only natural abundance of  $N^{15}$  in the  $\beta$  position, but when 80 percent of a sample of diazonium salt was decomposed in water, degradation of the remaining 20 percent showed 2.6 percent  $N^{15}$  in the  $\beta$  position, 2.2 percent above natural abundance. Within the precision of the mass spectrometric analysis, this result was independent of the temperature of the decomposition. It is clear that the reaction



occurs to a significant extent during the hydrolysis.

The same reaction also occurs to a somewhat greater extent with *p*-toluenediazonium ion. The layer extent of reaction does not appear to be enough to account for on the basis of a familiar 1,2 shift, for the migratory aptitude of the *p*-tolyl group is usually much greater than that of the phenyl group.

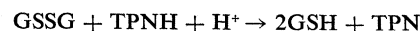
These results are consistent with an intermediate with nonequivalent but readily interconvertible nitrogen atoms common to both the isotopic rearrangement and the solvolysis, for which a reversibly formed intermediate was previously deduced from the acceleration of the rate by thiocyanate ion.

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### Red Blood Cell Glutathione Reductase in Gout

The rate of the reaction



catalyzed by glutathione reductase, was considerably increased in hemolyzates of red cells from six untreated individuals with elevated serum uric acid values and clinical histories of gouty arthritis. The rate of this reaction in red cell hemolyzates from four other persons who were taking probenecid for gout was normal. The mean reaction rate for hemolyzates from the six untreated gouty individuals was  $14.58 \times 10^{-9}$  moles of TPNH oxidized per milliliter per minute, referred to a hemoglobin concentration of 0.105 g percent. The mean reaction rate for red cell samples from 22 normal controls was  $10.72 \times 10^{-9}$ . Value of *p* is less than .01. Possible mechanisms whereby increased glutathione reductase activity might be involved in gout through increased production of ribose via the pentose phosphate pathway or through an effect on renal tubular permeability to uric acid will be discussed. (Grant HE-04516-04, USPHS)

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### Studies on the Function and Structure of a Multienzyme Complex

The *Escherichia coli* pyruvate dehydrogenation system is a multienzyme complex (molecular weight approximately 4.8 million) which catalyzes a multistep oxidative decarboxylation of pyruvate to produce acetyl coenzyme A,  $\text{CO}_2$ , and reduced diphosphopyridine nucleotide. The complex has been separated into three essential enzymatic components, (i) pyruvic carboxylase (183,000), (ii) dihydrolipoic dehydrogenase (112,000), and (iii) lipoic reductase-transacetylase (approximately 1.6 million). The latter component appears to be an aggregate of a subunit with a minimal molecular weight of approximately 30,000. Each molecule of the complex contains approximately 16 molecules of the carboxylase and 8 molecules of the dehydrogenase embedded in a matrix of the lipoic reductase-transacetylase.

This pyruvate dehydrogenation complex provides a unique opportunity to correlate functional properties as revealed by biochemical analysis, with ultrastructure as revealed by electron microscopy. The picture of the struc-

tural organization of the complex which has emerged from biochemical studies, that is, an organized mosaic of enzymes in which each of the component enzymes is uniquely located to permit efficient implementation of a consecutive reaction sequence, has been confirmed and extended by electron microscope studies (Fernandez-Moran).

Electron micrographs of the complex negatively stained with phosphotungstate show a square or, in some cases, a rectangular structure (approximately  $250 \times 320 \text{ \AA}$ ) featuring an orderly array of subunits or "knobs." These "knobs" are also seen in micrographs of the complex stained with uranyl acetate. A tentative correlation of many of these "knobs" and the pyruvic carboxylase component of the complex has been made. It is anticipated that these correlative studies will ultimately provide a detailed picture of the structural organization of this multienzyme complex at the molecular level.

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#### **Inhibitory Effects of**

#### **p-Fluorophenylalanine and Puromycin on the Induction of Thymidine Kinase by Vaccinia-Infected Mouse Fibroblasts**

Thymidine kinase activity of suspension cultures of L-M mouse fibroblasts is markedly enhanced following vaccinia infection. The augmentation of enzyme activity can be detected at 1.5 hours postinoculation (PI) of virus and increases to a maximum at about 5–7 hours PI. This corresponds to the time of active viral DNA synthesis. Experimental evidence suggests that the induced enzyme represents a new protein, possibly coded by the vaccinia genome. A bromodeoxyuridine resistant subline of L-M cells has been selected which lacks the enzyme, thymidine kinase. The enzyme is produced, however, after the resistant cells are infected with vaccinia. Moreover, inhibitors of protein synthesis prevent the induction of thymidine kinase. After L-M cells are incubated with  $5 \times 10^{-5}M$  puromycin·HCl or with  $200 \mu\text{g/ml}$  of p-fluorophenylalanine (FPA) for 4 hours, the incorporation from the fourth to the fifth hour of DL-tryptophane- $2\text{-C}^{14}$  into cell proteins is inhibited by 62 percent and 33 percent respectively. The FPA inhibition can be prevented by adding  $300 \mu\text{g/ml}$  of L-phenylalanine to the medium. High concentrations of FPA and puromycin·HCl do not inhibit

thymidine kinase activity, per se. However, at the concentrations of FPA or puromycin·HCl, used above, thymidine kinase induction by vaccinia is completely prevented. Inhibition of enzyme induction by FPA can also be prevented by L-phenylalanine. If puromycin·HCl or FPA is added to cell media at 3 hours after inoculation of vaccinia, the initial augmentation of thymidine kinase activity ceases. If L-phenylalanine is added to the FPA inhibited cells at 6 hours PI, thymidine kinase induction is resumed.

SAUL KIT, D. R. DUBBS

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#### **Gossyverdurin: A New Pigment Found in Cottonseed Pigment Glands**

The constituents of cottonseed pigment glands were fractionated by column chromatography and a new green pigment was isolated. The new pigment, which was named gossyverdurin, was found to be structurally related to gossypol which is yellow in color and much more stable than the new compound. Gossyverdurin reacts with p-anisidine under the conditions used for the determination of gossypol to give an absorption peak similar to that obtained with gossypol. In addition a second absorption peak appears at  $342 m\mu$ . Gossyverdurin contains nitrogen which is liberated on acid hydrolysis in the form of ammonia.

Acute oral toxicity tests with rats showed that gossyverdurin is more toxic than gossypol and the gossypol-related compounds, diaminogossypol and gossypurpurin.

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#### **On the Optical Forms**

#### **of the Brightest**

#### **Extragalactic Radio Sources**

A total of 45 galaxies whose identifications as radio sources are of high quality have satisfactory photographic plate material at the Mount Wilson-Palomar Observatories and California Institute of Technology. This material was used for a study of the form characteristics of extragalactic radio sources.

The radio sources can be approximately separated into 38 "strong" and 7 "weak" sources, on the basis of their luminosity and photographic appearance. Most of the "strong" sources are of an elliptical type with

greatly extended envelopes of varying importance—and having, in some cases, double or multiple nuclei; three may be limiting cases of the above, in which the envelopes are insignificant in importance.

The "weak" sources as a group are spiral galaxies. The boundary in luminosity that separates the "strong" and "weak" sources is near  $10^{40}$  erg/sec radiated power.

There is an absence of highly flattened systems among the "strong" radio sources, which appears to be significant; the "strong" group is sharply differentiated from the "weak" radio sources in this respect. The only qualification to this conclusion would be if radio radiation were emitted preferentially from the "strong" sources in a polar direction. This possibility seems unlikely, from the observed radio structure of extragalactic sources. (Supported by contracts between the Office of Naval Research, and California Institute of Technology, and the University of Chicago)

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#### **Internal Structures and Mode of Uplift of Texas and Louisiana Salt Domes**

Detailed studies of salt layering, and petrofabric studies of salt and anhydrite, as well as shape orientation of salt crystals in the Morton Salt Company Mine, Grand Saline salt dome, Texas, support the thesis that this dome grew differentially upward in pencils and spines rather than moving uniformly upwards as an entity. Ground water solution removed the uppermost part of the dome and left only the internal salt structures as clues to the mode of uplift.

Published salt structure maps from mines in Winnfield, Weeks Island, and Jefferson Island salt domes, Louisiana, can be interpreted to conform to the mode of uplift postulated for Grand Saline salt dome. The salt mines in Jefferson Island and Weeks Island domes appear to be structurally high cross sections of the domes, as shown by complex disharmonic structures, topographic highs over the domes (indicating recent uplift), and lack of cap rock development. The salt mines in Winnfield and Grand Saline salt domes are cross sections that are structurally lower (deeper) as shown

by broad scale folding, topographic depressions over the domes, and thick cap rock development. Unpublished mapping of the few visible structures in the mine in Hockley salt dome, Texas, suggests that it is also a structurally low cross section.

Stratigraphic data from the margins of other Texas and Louisiana salt domes show that the structurally high regions of growing salt domes have migrated in position through time, thus suggesting that the thesis of local differential uplift can be extended to include most, if not all, salt domes of this region.

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### Control Mechanism for Bicarbonate Incorporation into Aspartic Acid

Over a decade ago growth studies with *Lactobacillus arabinosus* gave evidence for a role of biotin in the carboxylation of pyruvic acid, and it was demonstrated that the incorporation of bicarbonate into aspartic acid in intact cells was inhibited by aspartic acid. In the present investigation, the biosynthesis of aspartic acid has been demonstrated in toluene-treated cells and in cell-free extracts using bicarbonate, pyruvic acid, and glutamic acid as the substrates. Adenosine triphosphate,  $Mg^{++}$  or  $Mn^{++}$ , and pyridoxal phosphate are also required, and the reaction is inhibited by avidin. Inorganic phosphate exchange with adenine triphosphate catalyzed by the system is dependent upon the presence of adenine diphosphate and bicarbonate, which is characteristic of other recently studied biotin enzymes catalyzing bicarbonate incorporation.

Aspartic acid inhibits the incorporation of bicarbonate in intact cells utilizing glucose as a source of energy as well as a source of pyruvate. In contrast, aspartic acid does not affect the utilization of bicarbonate in either toluene-treated cells or in cell-free extracts, even when glucose is used as a source of pyruvate in the toluene-treated cells. Thus, it appears that in intact cells aspartic acid inhibits the transport of bicarbonate to the active site on the carboxylation enzyme. This represents an unusual type of control mechanism in which the end-product controls the transport of one of the substrates required for its synthesis. Such control mechanisms may be of general metabolic significance.

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### Structure, Morphology and Color of Colloidal Gold

Gold hydrosols, prepared by reduction of chlorauric acid with sodium citrate, citric acid, hydrogen peroxide, hydrazine hydrate, hydroxylamine hydrochloride, and phosphorus in ether, were examined by the methods of electron microscopy, x-ray diffraction line broadening, and light scattering (extinction coefficients, depolarization, and absolute light scattering coefficients). In contrast to much of the earlier work, all measurements were carried out on identical samples.

Measured extinction coefficients for gold hydrosols containing particles of a uniform size and of a shape corresponding to a cube-octahedron show close agreement with the theoretical results of Mie for conducting spheres. The presence of "trigons" resulted in a broadening of the absorption peak and a shift to a higher wavelength. The experimental absorption curves fell between the theoretical curve of Mie and that of Gans for prolate ellipsoids of axial ratio 0.77.

A plot of the depolarization factor,  $p_u$ , versus wavelength gave a characteristic curve with a maximum occurring at approximately 4000 Å and a sharp rise in the vicinity of 5500 Å. The values of  $p_u$  for sols containing a large number of "trigons" were approximately twice that of more uniform sols. In general, the observed curves and the theoretical curves of Gans for ellipsoids showed close agreement.

As would be expected, the average particle size values calculated from absolute light scattering coefficients using the equations of Mie were considerably larger than the values obtained from electron microscopy, especially for sols containing "trigon." Excellent correlation was observed between x-ray diffraction line broadening and electron microscopic measurements, with respect to both particle shape and size. Best agreement was obtained for sols of uniform particle size having an average diameter between 100 and 200 Å.

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### Nuclear States of $N^{14}$

Experiments have been carried out involving excited states of the compound nucleus  $N^{14*}$  in the 10- to 12-Mev range of excitation energies. These states can be formed by deuterons incident on  $C^{13}$  and protons incident on  $C^{13}$ . Experiments with particles in two channels with large partial

widths for formation of the states allow a fairly complete experimental determination of the state parameters. The states of  $N^{14}$  are interesting from the point of view of nuclear structure because a number of them may possibly be characterized by the motion of two nucleon holes in the stable nuclear shell structure of  $O^{16}$ . In addition to measurements of proton and deuteron elastic cross sections, our work consisted in the measurement of a number of reaction cross sections, notably the total cross sections for the production of neutrons by protons and deuterons from the radioactivity of  $N^{13}$  produced in the  $C^{13}$  and  $C^{13}$  targets. Using the nuclear dispersion theory of Wigner, Breit, and collaborators in the single level approximation, we have attempted to fit the elastic scattering and total cross section data with reasonable choice of parameters. Positions, total widths, angular momenta, parities, and the principal partial widths have been determined for a considerable number of states.

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### Structure and Function in Mammalian Hemoglobins

Mammalian hemoglobins differ in oxygen affinity and in their sensitivity to pH and  $CO_2$ . These differences are associated with the metabolic requirements of the animals. In general, the smaller the animal, the higher is its metabolic rate, and the higher is the pressure at which oxygen is delivered to the tissues by the hemoglobin. This molecular adaptation appears to be augmented by differences in the ratio of acidic to basic amino acids in the molecule. A comparison of the amino acid composition of a number of mammalian hemoglobins shows that variation in the content of the acidic amino acids and their amides appears primarily responsible for the differences in isoelectric point of the proteins. The basic amino acid content is relatively constant. An examination of data in the literature on the amino acid composition of several other homologous series of proteins (bacterial cytochromes, lactic dehydrogenases, chymotrypsins, trypsinogens, and vertebrate skin gelatins) shows that the acidic amino acid content is much more variable than that of the basic amino acids. Examination of

one of the recently proposed genetic codes suggests that the tendency for basic amino acids to remain constant relative to the acidic amino acids may result from an intrinsic property of the code.

Although the exact code is still uncertain and the number of homologous series of proteins is very small, it seems, at present, unlikely that the phenomenon is accidental.

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### **Electromagnetic Two-Body Problem**

The circular motion of two classical point charges interacting through their time-symmetric electromagnetic fields is determined with complete relativistic rigor. Expressions are obtained for the total energy and angular momentum of this two-body system, which include the contributions of the field. This work is a preliminary to a calculation, undertaken jointly with J. Schlosser and now in progress, of the relativistic electromagnetic interaction of two classical particles with charge, spin, and magnetic moment, and of the quantization of this classical two-body system.

The purpose of the investigation is to check the possibility that some elementary particles may be atoms composed of other elementary particles in states where the principal interaction is due, not to the Coulomb force between point charges, but to forces where the intrinsic magnetic moments of the particles play an essential role and must not be considered as small perturbations. A very rough, but very simple, estimate by H. C. Corben indicates that this possibility is worth exploring. (Sponsored by the Office of Aerospace Research, U.S. Air Force)

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### **Relation of Antigen Dosage to the Forssman Hemolysin Response in Rabbits**

Regression curves, relating peak antibody titer, length of the latent period, and rate of serum antibody rise to log antigen dose of sheep red cells injected intravenously, have been constructed over a 4-million dose range for the initial response and over a 2.5-billion range for the anamnestic response.

As antigen is increased, more antibody is formed in less time. On

the linear portion of the curves of both responses, regression coefficients for the following pairs of measures are of approximately the same magnitude, but are opposite in sign: log peak titer and length of the latent period; rate of serum rise to peak titer and the elapsed time of the rise. The results suggest the shortening of one or more of the following events taking place during the latent period: induction, generation time, and maturation of the antibody-forming cells. As regression lines approach the asymptote of maximum antibody production, the rise of serum antibody to peak titer changes from a single rate to a series of decreasing rates. The anamnestic, as compared to the initial response, occurs after the injection of less antigen (16 versus 10,000 red cells), is faster, and reaches a level 20 times higher after a dose of 1000 cells, although it is only slightly higher at the upper asymptote. (Supported by the U.S. Atomic Energy Commission)

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### **Importance of Calcium Homeostasis and Its Control by the Parathyroids**

Calcium is the heaviest element found in abundance in vertebrate and many invertebrate animals. As a cation, its significant influence on important biochemical reactions has long been known, although the mechanism is not completely understood. Abundantly supplied by nature, this element must be carefully conserved by vertebrates because of the low solubility of most of its salts; in fact, its use is limited and indirectly controlled by relative insolubilities of its phosphate salts. Many physiological processes, including bone growth and remodeling, require that ionic calcium levels of circulating fluids be closely regulated. This is accomplished by a feed-back system whereby decreases in plasma calcium ion concentrations trigger increased release of parathyroid hormone. The hormone, in turn, causes replacement of calcium lost by releasing the ion from bone back into body fluids. The intricacies of this control are necessitated by a condition of undersaturation of body fluids in regard to secondary calcium phosphate and the fact that this fluid immediately assumes a state of supersaturation when it comes in contact with tertiary calcium phosphate in its crystal form in bone. Work from this laboratory has been concerned with the interplay between the simple equilibration of

fluid and solid phases and the biochemical processes opposing this equilibrium. The metabolic energy required for this opposition is thus utilized in the regulation of biochemical processes by ionic calcium levels.

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### **Molecular Packing and Crystal Structure of Aliphatic Carboxylic Acids**

If one plots the melting points of the normal fatty acids versus the number of carbon atoms in the aliphatic chain, the values fall on two smooth curves. Furthermore, these curves are not monotonic, but exhibit a minimum value in the region of five carbon atoms.

It is thus rather interesting to compare the solid state structures of these acids in the low molecular weight region, namely those containing from one to seven carbon atoms. Since all of these acids are liquid at room temperature, it has been only recently that their crystal structures have been investigated. At present the structures of those molecules containing five or fewer carbon atoms have been investigated by low-temperature x-ray diffraction techniques. It is the purpose of this paper to describe the structural features of these acids as well as those of the related substances, acrylic acid and the bicarbonate ion.

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### **Infrared Spectra by Matrix Isolation of Lithium Fluoride, Lithium Chloride, and Sodium Fluoride**

The matrix isolation method, which was adapted for infrared spectroscopy of molecular species existing at high temperatures by Linevsky, has been employed to examine the spectra of LiF, LiCl, and NaF. Rather complex spectra were found. Most of the features of these spectra must arise from polymeric alkali halide species. Spectra were taken for Ar, Kr, and Xe matrices and the effects of matrix material are considered. Isotopes effects as well as spectral shifts after matrix diffusion assist in the interpretation of these data.

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### Sizable Nuclear Polarization of He<sup>3</sup> Gas

We wish to report here the achievement of nuclear polarization in excess of 20 percent in He<sup>3</sup> gas at 1 mm-Hg pressure. The polarization is accomplished by means of a process involving metastability exchange with optically pumped helium atoms in the 2<sup>3</sup>S<sub>1</sub> state. Details of the process have been reported elsewhere. [G. K. Walters, F. D. Colegrove, L. D. Schearer, *Phys. Rev. Letters* **8**, 439 (1962)]. The factor presently limiting the polarization to about 20 percent is the insufficient pumping light intensity from our 100-watt helium discharge lamp; hence, considerable polarization enhancement should be possible. Even with the presently obtainable 20-percent polarization, however, important nuclear scattering experiments involving spin-dependent interactions become possible.

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### Kinetics of Production of Schizokinen, a Bacterial Division Control Factor

The duration of division lag in microbial cultures often is an inverse function of inoculum density. It has been postulated that cell division requires accumulation to critical concentration of an endogenous "diffusible factor." This concept was investigated in cultures of *Bacillus megaterium* in a mineral salts-sucrose medium, into which this organism releases a potent "division activator," designated a *schizokinen* (an "activity unit" of which effects lag time reduction equivalent to a standard).

Bioassays of culture filtrates demonstrate that extracellular schizokinen accumulates exponentially until the first cell division; thereafter net secretion diminishes or ceases until well into the exponential phase. These data indicate also that schizokinen concentration per cell at the time of the first division is an inverse function of the logarithm of inoculum concentration over a 10<sup>4</sup>-fold range of inoculum size. Hence, schizokinen attains a "threshold" concentration range (3.2 to 18 units per milliliter of medium, for inocula of 30 to 450,000 cells) prior to the first division. When filtrate is added with inoculum, a minimum effective concentration is required to reduce lag 1 hour below controls (0.1 to 8.4 units schizokinen per milliliter,

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for 2 to 200,000 cells). This evidence does not distinguish between a requirement for extracellular or intracellular schizokinen at critical concentration.

A mathematical description of schizokinen concentration per cell as a function of time during lag in cultures of large inocula was developed and found to be valid for prediction of lag in cultures of small inocula.

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### Ascorbic Acid in the Nutrition of Plant-Feeding Insects

Efforts to devise diets for rearing plant-feeding insects have not been very successful. Apparently, these insects need one or more chemically unstable nutrients because fresh plant food is often a necessity. In previous studies the need for dietary ascorbic acid, a very unstable substance, was reported for several species of insects. Additional work with the bollworm, *Heliothis zea* Boddie, disclosed the importance of ascorbic acid at various stages of the life cycle. Insects fed defined diets containing different concentrations of ascorbic acid were analyzed for their content of this vitamin. An absolute dietary requirement for the vitamin was proved, since it was absent in the tissues of insects whose diets contained no vitamin. Bollworms collected in the field contained a higher concentration of vitamin than those reared in the laboratory tests. The high concentration of vitamin found in the eggs was particularly interesting. Not all insects feeding on plants need dietary ascorbic acid. Tissues of the pink bollworm, *Pectinophora gossypiella* Saunders, contained ascorbic acid even though the larvae were fed a diet free of ascorbic acid. The effect of ascorbic acid on insect metabolism is being investigated.

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### Control of Dimorphism in the Female Honeybee

The female egg of the honeybee can develop into either a queen or a worker, and the direction of differentiation depends primarily, if not entirely, on the food received during larval life. Queen bees can be reared in an incubator by feeding larvae on royal jelly removed from the cells of queen larvae that are being fed by bees in the hive.

Compounds in royal jelly are labile, and during storage royal jelly loses its ability to produce queens, and ultimately, to support larval growth. Royal jelly has been fractionated, and a fraction that is necessary for the differentiation of queens has been separated. When the active fraction is omitted from the diet, but all other fractions are recombined, only normal workers, or slightly queen-like workers, are produced. When all fractions are recombined the same percentage of larvae develop into queens as develop on whole, fresh royal jelly under the same conditions of feeding. The nature of the action of the active fraction is not understood. Its absence seems to destroy the growth or differentiation regulating mechanism in some of the larvae. The same effect has not been obtained in experiments on the balance of nutrients.

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### New Approach to Hypervelocity Theory

Based on the assumption that a fluid state exists behind the shock wave which is created by the high velocity impact of the projectile and target, an expression is developed for a penetration parameter as a function of an impact velocity parameter. This formula is obtained from the simultaneous solution of one-dimensional equations of a viscous compressible fluid. The two constants in the theoretical formula are determined from the experimental data for identical projectile and target materials. One of the constants is found to be the same value for all the materials under consideration, that is, steel, aluminum, copper, tin, and lead. The other constant is the ratio of dilatational wave velocity of materials at standard conditions and the dilatational wave velocity of the materials in the fluid state. This constant is found to be a function of dilatational wave velocity of materials at standard conditions. For a given impact velocity and the corresponding dilatational wave velocity of the material at standard conditions the penetration parameter can be readily determined. The agreement between a single theoretical curve, plotted in a penetration parameter versus the impact velocity parameter, and available experimental data for six different materials, is surprisingly good.

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