Table 1. The incorporation of activity into the amino acids in the hydrolysates of the "peptide" fraction after 2—^{14}C-pyruvate infiltration into *Aspl. septentrionale*.

Time, h	½	1	2	24
Glutamic, cpm	140	493	266	60
Alanine, cpm	243	175	94	
γ-Aminobutyric, cpm	94	163	—	—
Valine, cpm	15	—	—	—

and α-aminobutyric acid were found labelled in this fraction. Alanine was the major labelled compound after half an hour, but already after one hour the activity of glutamic acid was greater (Table 1). Whereas alanine decreased from the "peptide" fraction it increased in the proteins. However, the activity of compounds 5, 6, 7, 8 and 11 also had a peak within 10 to 30 min when 2—^{14}C-pyruvate alone was infiltrated. Glutamine was not labelled in this case. Inactive glycine, when infiltrated together with active pyruvate, caused a great inhibition of the labelling of these compounds during the first minutes after infiltration, whereafter the activity of compounds 6 and 7 as well as that of glutamic acid greatly increased, showing again the relationship of compound 6 to glutamic acid. In this case even glutamine was slightly labelled after 3 h. Glycine had only a slight inhibitory effect to the alanine containing compounds 5 and 8, while the activity of alanine itself increased due to the greater amount of nitrogen available for transamination reactions.

1. Linko, P. and Virtanen, A. I. *Acta Chem. Scand.* **12** (1958) 68.
2. Leloir, L. F. and Cardini, C. E. *Biochim. et Biophys. Acta* **12** (1953) 15.
3. Pogell, B. M. *Biochim. et Biophys. Acta* **21** (1956) 204.
4. Linko, P., Holm-Hansen, O., Bassham, J. A. and Calvin, M. *J. Exptl. Botany* **8** (1957) 147.
5. Virtanen, A. I. and Miettinen, J. K. *Biochim. et Biophys. Acta* **12** (1953) 181.

Received September 16, 1957.

The Influence of Anharmonicity upon the Vibrational Probability Density and Mean Amplitudes in Diatomic Molecules

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The influence of anharmonicity upon the vibrational probability density and mean amplitudes in diatomic molecules has been investigated by using two different models to describe the molecular vibrations:

a) an anharmonic oscillator with perturbation terms of first and second order in the potential function,

b) an anharmonic oscillator described by a Morse potential function.

For case a) the probability density $w(r)$, where $r = R - R_e$ is the deviation of the interatomic distance R from its equilibrium value R_e, has been calculated by two different methods. In the first place, an expression for $w(r)$ has been developed by a superposition of probability densities for the different stationary states, according to the equation

$$w(r) = \sum_v |\psi_v(r)|^2 \exp(-E_v/kT) / \sum_v \exp(-E_v/kT)$$

where E_v and ψ_v are the corresponding eigenvalues and normalized eigenfunctions for the vibrational state v, k is Boltzmann's constant, and T is the absolute temperature. The eigenvalues E_v and eigenfunctions ψ_v have been obtained by a second order perturbation calculation similar to that performed by Hutchisson [1] (the results given by this author are, however, somewhat erroneous), and the sums have been evaluated by using well known properties of the Hermite orthogonal functions.

In the second place, the probability density $w(r)$ has been calculated by making use of an analogy to the solution of the time-dependent Schrödinger equation for the system (compare Bloch [2] and Kennard [3]).

In both cases the calculations are somewhat tedious, the result is, however, quite simple:

$$w(r) = (a_0 + a_1 r + a_2 r^2 + a_3 r^3 + a_4 r^4 + a_6 r^6) w^\circ(r)$$

where $w^\circ(r)$ is the probability density for the harmonic case, and the a_k's are explicitly given as functions of the temperature T, the vibrational frequency ω_e for the corresponding harmonic oscillator, and the spectroscopic anharmonicity constant x_e.

If the most probable value of R is denoted by R_m, and the values of R corresponding to a probability density equal to one half of the maximum value are denoted by $R_{h,1}$ and $R_{h,2}$, it is found that to terms of second order

$$R_{h,1} - R_m = r_h^\circ(1 + x_e^{1/2}b_1 + x_e b_2),$$
$$R_m - R_{h,2} = r_h^\circ(1 - x_e^{1/2}b_1 + x_e b_2),$$

where r_h° is the half width for the harmonic case, and the b_k's are functions of the temperature T and the vibrational frequency ω_e. The probability density curve is, as it is seen, unsymmetric, the unsymmetry appearing already in a first order treatment.

Furthermore, explicit expressions have been developed for the mean amplitudes

$$u_e = [\overline{(R-R_e)^2}]^{1/2},$$
$$u_a = [\overline{(R-\bar{R})^2}]^{1/2},$$
$$u_m = [\overline{(R-R_m)^2}]^{1/2},$$

where the reference values of the interatomic distance in the three cases are the equilibrium value R_e, the mean value $\bar{R}$, and the most probable value R_m, respectively.

In case b) of the Morse-potential approximation the calculations have been carried out in a straightforward manner, considering only the lower vibrational states. However, the numerical results obtained by the two methods seem to be in very good agreement, as indicated by the values for the hydrogen molecule listed in Table 1. The necessary spectroscopic data have been obtained from Herzberg [4]. The values obtained when assuming harmonic vibrations are also given.

Table 1. Numerical data for hydrogen in Å units at $T = 300$ °K.

	Harm. osc.	Pert. meth.	Morse pot.
u_e	0.0872	0.0915	0.0917
u_a	0.0872	0.0890	0.0891
u_m	0.0872	0.0893	0.0894
$R_{h,1}-R_m$	0.1027	0.1075	0.1075
$R_m-R_{h,2}$	0.1027	0.1009	0.1008

A more detailed report on the calculations performed is to be published in *Kgl. Norske Videnskab. Selskab Skrifter.*

1. Hutchisson, E. *Phys. Rev.* **37** (1931) 45.
2. Bloch, F. *Z. Physik* **74** (1932) 295.
3. Kennard, E. H. *Z. Physik* **44** (1927) 326.
4. Herzberg, G. *Molecular Spectra and Molecular Structure*, Vol. I, *Spectra of Diatomic Molecules*, 2. Ed. Nostrand, New York 1950.

Received October 29, 1957.

N-Terminal Amino Acids of Human Pituitary Growth Hormone

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Analyses of the N-terminal amino acids of growth hormone prepared from ox pituitaries have been carried out by means of two methods. The dinitrofluorobenzene-technique (DNFB) of Sanger [1] has been applied by Li [2], who found two amino acids (phenylalanine and alanine) in equal molar proportions. These results were confirmed by the phenylthiohydantoin (PTH) method of Edman [3]. The latter method proved to be better since it was clearly demonstrable that the two amino acids were present as one mole each per mole of protein hormone [4].

This suggests a constitution formula of the ox hormone of two peptide chains linked together by SH-groups and ending in phenylalanine and alanine as N-terminal amino acids.

The following experiments were carried out to analyse the N-terminal amino acid of human pituitary growth hormone [5].

Material and methods. Growth hormone was prepared from human pituitaries by means of a method of Li and Papkoff [6]. 4 to 10 mg samples of the protein hormone were dissolved in 0.5—2.0 ml of phosphate buffer pH 9.0, ionic strength 0.1. An equal volume of 0.5 % phenylthio*iso*cyanate (Eastman Kodak) in ethanol was added and the mixture stirred for 2 h. The pH was maintained at 8.8 by the addition of small portions of 0.01 N sodium