

readily obtained. In practice, S is given such a fixed value (which need not be known precisely) that R will be several hundred ohms, or more, for either balance. Under these conditions, the resistance of the cell E will be quite negligible and

$$\frac{r_1}{r_2} = \frac{R_2 + S}{R_1 + S}$$

This principle has, obviously, other applications; for example, to thermo-couple measurements, in view of the low values possible for the resistances of S and G .

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Decomposition of Hydrogen Peroxide by Catalase

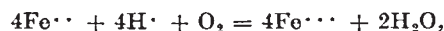
IN a recent letter¹ Johnson and van Schouwenburg dispute Keilin and Hartree's observation² that catalase activity depends on the presence of oxygen. In their reply, Keilin and Hartree³ point out that the experiments of Johnson and van Schouwenburg are only qualitative and therefore not conclusive.

Soon after the publication of Keilin and Hartree's paper², we attempted to repeat their experiments, following all their directions as closely as possible, but with negative results. Our experiments were carried out in Warburg manometers with a purified catalase from red blood cells and with an enzyme from cucumber seeds prepared according to Zeile⁴. To ensure complete absence of oxygen, the manometers were gassed with purified nitrogen until no oxygen could be detected by the ferrous pyrophosphate method in control experiments. No difference was found in the rate of the decomposition of hydrogen peroxide in presence or absence of oxygen. The non-enzymatic decomposition was controlled in experiments with heat-inactivated enzyme and was certainly not larger than in the experiments of Keilin and Hartree.

The successive oxidation and reduction of catalase was first suggested by Haber and Willstätter⁵ so long ago as 1931. This theory has been developed by one of us⁶ along the lines previously suggested by Haber and Weiss⁷. It is based on a radical chain mechanism: the reduction of the ferric form of catalase is brought about by the anion of hydrogen peroxide (HO_2^-) and the oxidation of the ferrous form by the hydrogen peroxide molecule itself. The decomposition of hydrogen peroxide would therefore proceed with the same velocity in the presence and absence of oxygen, as we have indeed observed.

Although, contrary to Oppenheimer and Stern⁸, it cannot be generally assumed that an oxidation by hydrogen peroxide would be faster than an oxidation by molecular oxygen, a re-oxidation of reduced catalase by oxygen only would have the effect that

no decomposition of hydrogen peroxide could take place at all. The reason for this is that, unless a radical chain mechanism is assumed, all the oxygen formed by the reduction of the ferric form of catalase must be quantitatively used up again for the re-oxidation of a stoichiometric amount of the ferrous form and the oxygen thereby reduced to hydrogen peroxide. To avoid this difficulty, Keilin and Hartree formulated the oxidation reaction by the following equation:



thereby assuming the reduction of oxygen to water without the intermediate formation of hydrogen peroxide. This is contrary to accepted theories of autoxidation and would appear impossible on the basis of the kinetic theory if only for the reason that it implies collisions of a very high order.

Further, on the basis of Keilin and Hartree's theory, one would expect the reaction, if carried out in nitrogen, to show the characteristic curve of an autocatalytic process owing to the increasing formation of oxygen. This is, however, not corroborated by the figures of Keilin and Hartree.

Conclusions as to the behaviour of catalase derived by analogy from spectroscopic observations on so-called 'azide- or hydroxylamine-catalase' cannot be accepted as strong evidence for the problem under discussion.

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¹ Johnson, H., and van Schouwenburg, K. L., *NATURE*, **144**, 634 (1939).

² Keilin, D., and Hartree, E. F., *Proc. Roy. Soc., B*, **124**, 397 (1938).

³ Keilin, D., and Hartree, E. F., *NATURE* **144**, 787 (1939).

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⁵ Haber, F., and Willstätter, R., *Ber. dtsh. chem. Ges.*, **64**, 2344 (1931).

⁶ Weiss, J., *J. Phys. Chem.*, **41**, 1107 (1937).

⁷ Haber, F., and Weiss, J., *Proc. Roy. Soc., A*, **147**, 332 (1934).

⁸ Oppenheimer, C., and Stern, K. G., "Biological Oxidations" (The Hague: W. Junk, 1939).

Osmotic Behaviour of *Palæmonetes varians* (Leach)

Palæmonetes varians var. *microgenitor* is a common brackish-water inhabitant of northern and western Europe well known for its ability to live in different concentrations of sea water. Specimens collected from brackish waters in the vicinity of Plymouth (salinity 23–29 ‰) are quite at home in the tanks of the laboratory containing normal sea water of salinity 34–35 ‰. Estimations of osmotic pressure of blood, by Baldes' modification of Hill's thermo-electric technique^{1,2}, from specimens kept in different concentrations of sea water, indicate that while this species is hypertonic in sea water of lower dilutions, it is definitely hypotonic in normal sea water.

The blood of animals that have been in the sea-water tanks for some months is isotonic with a solution of 2.3–2.1 per cent sodium chloride. Hypotonicity is maintained by the shrimps in dilutions of sea water equivalent up to about 2.0 per cent sodium chloride, when the internal and external media are observed to be approximately isosmotic (see accompanying graph). In sea water of 1.8–0.6 per cent sodium chloride, there is a slight fall in the value