

effect in concentrated solutions appears to be due to masking by unchanged material.

Fig. 3 shows the time course for one of our tested substances, adenylic acid. Exposure to ultraviolet light for 4 hr results in approximately 20% reduction in absorption at 260 m μ , whereas for 6 hr the reduction is approximately 40%. After 12 hr exposure, the selective absorption spectrum disappears. Uric acid, which has two peaks, one at 235 m μ and the other at 290 m μ , also behaves as do all the other nucleic acid derivatives. Complete obliteration of its absorption curve is observed in Fig. 4.

The curves showing the effect on the other substances studied are omitted for lack of space. It suffices to say that in all cases the curves are essentially similar and reproducible.

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Contact Potentials of Evaporated Iron Films in Air and in Nitrogen at Low Pressure

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Curves showing the variation of contact potential with time for metal surfaces prepared by the evaporation process have been obtained *in vacuo* at room temperature using the method of Zisman (1). A platinum plate is used as the standard of reference.

The change in potential of an iron surface amounts to several tenths of a volt and is due largely to the sorption of oxygen. The rate is pressure-sensitive between 0.01 and 10 μ , being very low and very high, respectively. The curves at 0.1 μ are characterized by a sharp rise, followed by a rounded peak, which falls to a level about 0.3 v below the maximum.

From between 0.01 μ and 0.1 μ to the higher pressures, the potential change is reversible upon alternating the pressure from high to low values. The trend of the variations indicates that irreversible sorption is continually taking place, also, and at a rate that is more or less dependent on the average pressure. The desorption rate is lower than the sorption rate.

When pure, dry nitrogen is introduced into the vacuum chamber, no appreciable change in surface potential with

time is observed, and pulsing of the pressure does not produce corresponding alternation in potential. This is assumed to indicate that most of the observed change is caused by oxygen.

Electrical resistance variations of similar evaporated films in the same vacuum range show that gross penetration of the oxygen and its combination with the iron are negligibly slow in comparison with the surface sorption effects, because the resistance increases at a negligible rate at pressures of 0.1 μ , whereas the surface potential curve reaches its maximum within a minute. A slight reduction in resistance of the iron films occurs within a few seconds after they are deposited and may be due to atomic rearrangement or to a drop in temperature.

Exposure to pressures of 10–100 μ resulted in slow and probably incomplete resistance changes, which are not reversible with reduction in pressure. At atmospheric pressure and humidity, evaporated iron layers up to 30 Å in thickness show little change in resistance after 30 min, but the resistance gradually increases over a period of hours at a rate depending on the thickness of the film.

In connection with these studies, experiments have been conducted in which evaporated metal films were produced within an electron microscope column. Electron diffraction patterns of these films were observed continuously from the time of deposition. It was possible to follow the formation of an oxide pattern over that of the metal as a function of time and pressure. For example, the first perceptible change at a pressure of 27 μ requires 1 1/2 min for iron, whereas 27 min were required for nickel.

Studies of the type described for iron have been made on a number of other metals, with results which vary considerably from metal to metal. Details will shortly be available.

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Improved Technique for Weighing Tissues with the Cartesian Diver

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In the course of experiments on respiration of embryonic chick hearts using the Cartesian diver technique, it became necessary to weigh the hearts. Ordinary methods with the analytic balance proved impractical. Zeuthen (1) describes a method for weighing tissues and cells in the Cartesian diver and presents equation (1) for calculation of weights:

$$RW_x = RW_{st} \left(\frac{1 - \frac{B}{B - p_x}}{1 - \frac{B}{B - p_{st}}} \right) \quad (1)$$

In this equation RW_x is the buoyed weight of the tissue, RW_{st} the buoyed weight of the standard, B is atmos-

TABLE 1
COMPARISON OF REDUCED WEIGHTS CALCULATED BY THREE EQUATIONS

Diver		Standard							Secondary standard							
No.	V_c in mm ³ equa- tion (5)	No.	Wt in air, mg	Sp. gr.	RW_{st} mg	p_e	p_{st}	No.	Actual weight				Calculated reduced weights			
									Wt in air, mg	Sp. gr.	RW_x mg	p_e	p_x	(1)	(2)	(6)
1G	227.7	1S	11.5	2.23	6.3	-378	-641	1F	970.9	1.018	17.2	-378	-1120	10.5	18.7	18.8
1G	227.1	2S	15.5	2.23	8.6	-406	-756	2F	693.9	1.007	4.8	-406	-612	7.1	5.0	4.8
1G	224.7	2S	15.5	2.23	8.6	-154	-517	2F	693.9	1.007	4.8	-154	-362	6.1	4.8	4.8
Mean	226.5															
2W	4.54	3S	0.690	2.23	0.381	-94	-861
2W	4.82	3S	0.690	2.23	0.381	+665	-116	3F	27.8	1.033	0.889	+665	-1014	3.051	0.901	0.887
2W	4.96	3S	0.690	2.23	0.381	+662	-99	4F	2.2	1.013	0.282	+662	+594	-2.428	.317	.296
Mean	4.77															
3W	5.14	3S	0.690	2.23	0.381	+153	-548
3W	5.22	3S	0.690	2.23	0.381	+132	-557
3W	5.16	3S	0.690	2.23	0.381	+691	-44
3W	4.87	3S	0.690	2.23	0.381	+717	-61	3F	27.8	1.033	0.889	+717	-913	5.23	0.874	.913
Mean	5.10															
Mean error														169%	4.7%	3.2%

pheric pressure or 10,000 mm Brodie's solution, p_x is the suspension pressure with the tissue on the diver, and p_{st} is the suspension pressure with the standard on the diver.

We constructed a diver suitable for measuring weights of embryonic chick hearts, but obtained implausible results with equation (1) when (a) the diver was not suspended at atmospheric pressure, and (b) when p_x was positive and p_{st} was negative in sign.

In the following equation, reliable results are obtained under all conditions ordinarily encountered:

$$RW_x = RW_{st} \left(\frac{1 - \frac{B + p_e}{B + p_x}}{1 - \frac{B + p_e}{B + p_{st}}} \right) \quad (2)$$

In equation (2), in addition to the symbols given above, p_e is the suspension pressure of the empty diver.

The method of Zeuthen requires that the diver be suspended successively with the standard weight, then with the unknown weight; the above modification requires, in addition, suspension of the empty diver. This procedure is rather laborious, although unavoidable under conditions in which air bubbles form on the outside of the diver; even then, successive suspensions must be made rapidly.

If the diver is exceptionally clean, if it has been immersed for several days, or if the suspension fluid contains minimum amounts of dissolved gases, air bubbles will not form on the outside of the diver, and a simpler procedure may be used.

This second procedure is based on the fact that under conditions of suspension, a diver contains a certain critical constant volume of gas, which may be called V_c .

If the suspension fluid is water with a specific gravity of 1.00, the following is true:

$$V_o B = (V_o + \Delta V)(B + p_e) = (V_o + \Delta V + RW_x)(B + p_x) \quad (3)$$

$$= (V_o + \Delta V + RW_{st})(B + p_{st}) \quad (4)$$

in which V_o is the volume of air in the diver at atmospheric pressure B , ($V_o + \Delta V$) is the critical constant volume V_c , and the other terms are as given above. Substituting and solving, the following equations may be obtained:

$$V_c = \frac{RW_{st}(B + p_{st})}{(p_e - p_{st})} \quad (5)$$

$$RW_x = V_c \left(\frac{B + p_e}{B + p_x} \right) - V_c \quad (6)$$

The diver is standardized as follows. The volume of air in the diver is adjusted until the diver is suspended, or floats upward, or sinks slowly at atmospheric pressure. The standard weight RW_{st} is placed on the diver, and the suspension pressure p_{st} is measured. The weight is shaken from the diver, and the suspension pressure p_e is measured. This is repeated several times, preferably with different original volumes, and the mean critical volume is calculated from equation (5).

Once standardized, the diver may be used for weighing tissues. The tissue is placed on the diver, the suspension pressure p_x is measured; the tissue is shaken from the diver, and the suspension pressure p_e is measured. The weight of the tissue is calculated from equation (6).

Results of weighings, using a gigantic diver and a diver suitable for weighing embryonic chick hearts, are shown in Table 1. It may be seen that there is a close check between actual weights and weights measured with the diver and calculated with equations (2) and (6), but poor checks between actual weights and weights measured with the diver and calculated with equation (1).

It is probable that under conditions where air bubbles

do not form rapidly on the outside of the diver the simplified method described can be applied to weighing tissues much smaller than embryonic hearts. The improvement in the equation given by Zeuthen makes accurate weighings possible under conditions where the diver is or is not suspended at atmospheric pressure.

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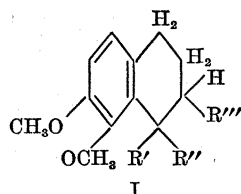
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On the Structure of Morphine and its Derivative Metopon

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Because the unique and valuable analgesic properties of the morphine derivative Metopon (methyldihydromorphinone) (4) may be due, in large measure, to the influence of the new methyl group, information regarding the latter's position in the molecule is of some moment. With a view toward elucidating this point, a method has been developed for degrading the more accessible dihydrothebaine (Metopon's precursor) to an optically active dimethoxytrialkyltetrahydronaphthalene Ia or Ib.



- (a) $R' = C_2H_5$; $R'' = C_2H_5$;
 $R''' = C_2H_5$
(b) $R' = CH_3$; $R'' = C_2H_5$;
 $R''' = C_2H_5$

Analysis. Calculated for $C_{18}H_{28}O_2$; C, 78.21; H, 10.21; OCH_3 , 22.46. Found: C, 78.22; H, 10.20; OCH_3 , 22.22. $[\alpha]_D^{20} - 52.7^\circ$ (c, 0.927, ethanol); $n_D^{20} = 1.5295$; $d_4^{20} = 1.027$; bp (evaporative distillation) 97–104° C/0.4 mm.

It is hoped to distinguish between the two possible isomers on the basis of results of synthetic experiments which are now nearly complete.

Degradation of Metopon according to this new scheme should lead to a methyl homologue of either Ia or Ib, distinguishable by synthetic processes.

Of equal and perhaps greater importance is the fact that this method of degradation now affords a complementary means of rigorously proving whether C^{13} is one of the points of closure of the heterocyclic nitrogen ring in morphine. The important synthetic approaches of Grewe (2, 3) and of Gates (1) both favor C^{13} as one point of ring closure; the other point, C^9 , appears well established.

Intimately linked with the above are the stereochemical implications associated with carbon atoms 9, 13, and 14 in the morphine molecule. All the foregoing will be fully reported in papers to be published elsewhere.

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Streaming Birefringence of Denatured Ovalbumin¹

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That the denaturation of globular proteins consists essentially of an unfolding of the polypeptide chain or chains is generally conceded, but evidence for the nature, or even existence, of the unfolded structure in solution is limited and indirect. Intrinsic viscosity data, which have been most used, are difficult to interpret since they yield only a measure of the effective hydrodynamic volume of the solute. Streaming birefringence, which yields essentially a measure of the particle length and is dependent only to a minor extent on molecular asymmetry and hydration, would appear to be a method of great potential value in the study of the denaturation process. Furthermore, results obtained over a wide range of velocity gradients provide an insight into the homogeneity (with regard to length) of the solute (1, 2).

The authors have completed more than 100 flow birefringence runs, in the concentric cylinder apparatus used by one of us previously (3), on ovalbumin denatured in various ways. The orientation angle χ (the angle between the optic axis and the direction of streaming) was measured, as well as the birefringence. The recently computed numerical solutions of Edsall and co-workers (8) for the flow orientation equations developed by Peterlin and Stuart (6, 7) were used for determining α (the ratio of the velocity gradient G to the rotary diffusion constant β) from the χ values obtained. The apparent length of the denatured ovalbumin molecules was calculated by applying the Perrin (5) theory for the case of an elongated ellipsoidal structure.

Denaturation of ovalbumin was carried out under such conditions that no precipitation, gelation, or appreciable turbidity occurred. Heat denaturation was studied in the pH range 1–4 using glycine-buffered and unbuffered solutions, and in the pH range 6–9 in the presence of veronal and phosphate buffers. Urea denaturation was followed in the pH range 6–9. The effect of cationic detergents on the acid side of the isoelectric point and of anionic detergents on the alkaline side was also examined. In a typical heat denaturation experiment 0.100 g of ovalbumin in 15.0 ml of buffer was heated at 100° C for a given period of time; the solution was cooled rapidly and diluted with 42.0 g of 95% glycerol. (The gly-

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