measuring capacitance between plate 5 and the combination, $\Delta C = 2.34$ μμf. (*c*) With plates 3 and 5, and all the other elements, connected together, and the capacitance measured between those two combinations, $\Delta C = 4.53$ μμf.

To determine the constancy of the cell calibration, measurements were made every few days over a period of about a month on the same sample of benzene. Each time a test was made the capacitance of the cell filled with air was first measured. Then the cell was filled with the benzene sample, and the capacitance was measured. Then the cell was emptied, and the residual benzene was evaporated by passing air through the cell. These tests were made with the connection described in (*c*). The values for ΔC obtained in this series of tests were all 4.53 ± 0.01 μμf.

No damage resulted to the cell when steam was passed through the jacket. This suggests that the cell can be used to measure dielectric constants over a considerable range of temperatures.

About 10 ml of liquid is required to fill the cell.

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Effect of Organic Solvents on the Emission Spectra of Sodium and Potassium in Serum and Aqueous Solutions [1, 2]

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A systematic investigation of solvents other than water for the dilution of serum for flame photometric determination of sodium and potassium has been made to discover a solvent which would eliminate some of the disadvantages of aqueous dilutions and possibly increase the range of measurement of small quantities of these elements.

Concentrations of 10–100% of anhydrous, sodium, and potassium free organic solvents in water with 0.3 mEq sodium and 0.01 mEq potassium added, were prepared for the study of the solvents' effect on the flame spectrum of sodium. The same dilutions of organic solvents containing 0.2% normal serum for a comparative study were prepared. Aqueous solutions of the same dilutions of normal serum and of sodium and potassium containing 0.02% Sterox[3] were used to represent 0% solvent. All sodium measurements were made with 0% solvent set at the 5% transmission value on a DU Beckman Spectrophotometer. Slit width was set at 0.05 mm (with sensitivity knob set near 0), oxygen pressure at 7 lbs, and acetylene at 3 lbs. All

[1] Paper read before the Southern California Section, American Association Clinical Chemists, Feb. 5, 1952, Los Angeles.

[2] Reviewed by the Veterans Administration and published with the approval of the chief medical director. The statements and conclusions of the authors are the result of their own study and do not necessarily reflect the opinion or policy of the Veterans Administration.

[3] Sterox, S. E., Monsanto Chemical Company, St. Louis, Mo.

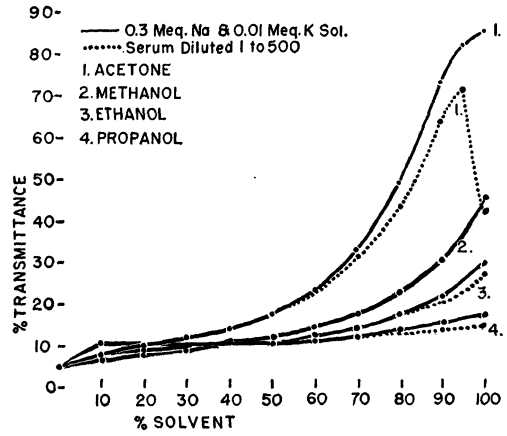

FIG. 1. Comparison of the effect of different concentrations of organic solvents on the flame spectra of sodium at 589.6 μ in diluted serum and in aqueous solutions, as measured with a Beckman Spectrophotometer with flame attachment #9200, acetylene-oxygen-burner assembly. Ordinate, relative light intensity read as % T on instrument's scale.

the above adjustments were kept constant, and the dark current only was adjusted for the slight differences of the flame background of the blanks for the different concentrations of the solvents. No measurable amounts of sodium or potassium were present in these blanks. Serum specimens were centrifuged to remove any precipitate which formed. It was observed (Fig. 1) that all the organic solvents enhanced the emission spectra of sodium progressively, especially above concentrations of 50%, and acetone gave much greater enhancement (seventeenfold) than the other solvents. Increasing amounts of protein were precipitated above 50% acetone, and loss of sodium with the precipitate was observed. Loss of the sodium salt with protein precipitate in 100% acetone removed more than 50% of the sodium from solution. Although serum protein was precipitated by 50% or more methanol, no loss of sodium with the precipitate was evident with this solvent. Of the solvents studied, only methanol exhibited this curious phenomenon. Little enhancement of the sodium spectra by propanol and isopropanol was observed.

The solvents were studied (Fig. 2), in the same manner as described for sodium, for their effect on the flame spectrum of potassium, using solutions containing 1.5 mEq sodium with 0.05 mEq potassium and solutions of normal serum diluted 100 times; 0% solvent was set at the 5% transmission value, slit width was fixed at 0.5 mm, oxygen pressure at 7 lbs, and acetylene at 5 lbs. The same relative order of effect of the solvents on intensifying the flame spectrum of potassium was observed as that for sodium. Acetone increased the flame spectrum of potassium thirteenfold. Loss of potassium occurred in all solvents above 50% concurrently with precipitation of serum proteins. Methanol solutions showed loss of potassium with the protein precipitate, although this solvent had shown no loss of sodium with the protein precipitate. Propanol and isopropanol showed little effect on the

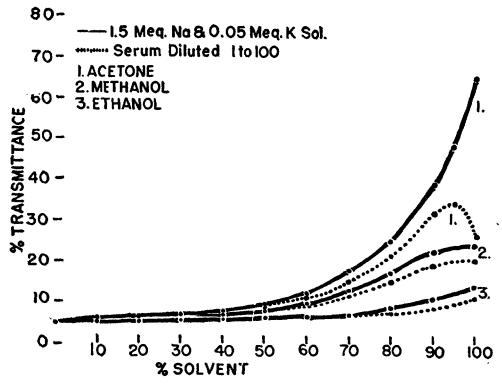

FIG. 2. Comparison of the effect of different concentrations of organic solvents on the flame spectra of potassium at 770.0 μ in diluted serum and aqueous solutions, as measured with a Beckman Spectrophotometer with flame attachment #9200, acetylene-oxygen-burner assembly. Ordinate, relative light intensity read as % T.

flame spectrum of potassium. All aqueous-organic solvent solutions of pure standards studied (Figs. 1 and 2) were not centrifuged and showed complete solubility of the sodium or potassium salts, except the 100% acetone solutions, which were turbid.

The enhancing effect of acetone on the flame spectrum of Na and K was also observed with the Weichselbaum-Varney (*1*) flame photometer.

The effect of protein on the flame spectrum of sodium and potassium was studied by using a 7.0% bovine albumin[4] solution which contained a trace of sodium and no potassium. This protein solution was diluted to 0.14% in 0–100% acetone containing 0.3 mEq sodium and 0.01 mEq potassium, for study of the flame spectra of sodium and 1.5 mEq sodium and 0.05 mEq potassium for the study of the flame spectra of potassium (Table 1). Acetone solutions containing 0.3 mEq Na and 0.01 mEq K with and without 0.014% bovine albumin read against an aqueous solution containing 0.3 mEq Na and 0.01 mEq K set at 5% T indicated that bovine albumin had no effect on the flame spectra of sodium (Table 1). Although bovine albumin was precipitated with 50–90% acetone, no loss of sodium with this precipitate occurred at these concentrations of solvent. No effect of bovine albumin on the flame spectrum of potassium (Table 1) was observed. Loss of potassium with bovine albumin precipitate occurred, beginning at 60% acetone.

TABLE 1

EFFECT OF ALBUMIN ON FLAME SPECTRA OF SODIUM AND POTASSIUM IN PRESENCE OF DIFFERENT CONCENTRATIONS OF ACETONE

Acetone (%)	Na spectra at 589.6 μ			K spectra at 770 μ		
	0.3 mEq Na 0.01 mEq K (% T)	0.3 mEq Na 0.01 mEq K 0.014% Albumin (% T)	0.014% Albumin (% T)	1.5 mEq Na 0.05 mEq K (% T)	1.5 mEq Na 0.05 mEq K 0.07% Albumin (% T)	0.07% Albumin (% T)
0	5.0	5.5	0.5	5.0	5.0	0.0
10	8.0	8.5	.5	5.0	5.0	.0
20	9.5	10.0	.5	6.0	6.0	.0
30	11.0	11.5	.5	7.5	7.5	.0
40	13.0	13.5	.5	9.0	9.0	.0
50	15.8	16.3	0.5	11.5	11.5	.0
60	18.5	19.5	1.0	15.0	15.0	.0
70	25.0	26.0	2.0	20.5	20.0	.0
80	34.0	35.0	2.0	29.0	28.0	.0
90	50.0	51.0	4.0	42.0	39.0	.0
95	59.0	58.0	3.0	53.0	43.0	.0
100	63.0	26.0	1.0	52.0	28.0	0.0

[4] Fraction V, Armour Laboratories, Chicago, Ill.

We have observed losses of sodium (7%) and potassium (10%) when trichloracetic acid is used to precipitate proteins from serum diluted 1/500 and 1/100. The use of trichloracetic acid (*2*) or other protein-precipitating agents to obtain protein-free filtrates for assay of Na and K by flame photometry is not recommended.

The observations reported here of the effect of a few common organic solvents on increasing the intensity of flame spectra from sodium and potassium have indicated the possibility of bringing more trace elements within the range of measurement of the flame photometer and the use of smaller samples for analyses. A further investigation is being made of the effect of other organic solvents, mixed organic solvents, and water on the emission spectra of sodium, potassium, calcium, magnesium, lead, and other elements.

References

1. WEICHSELBAUM, T. E., and VARNEY, P. L. *Proc. Soc. Exptl. Biol. Med.*, **71**, 570 (1949).
2. OVERMAN, R. R., and DAVIS, A. K. *J. Biol. Chem.*, **168**, 641 (1947).

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A Culture Technique for the Diagnosis of Infections with *Dermocystidium marinum* Mackin, Owen, and Collier in Oysters[1]

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A simple technique has been devised for the rapid and accurate diagnosis of infections with *Dermocystidium marinum* Mackin, Owen, and Collier, 1950 (*1*). This organism is now known to be widely distributed in oysters on the Gulf and South Atlantic coasts of the United States. Its pathological significance is discussed by Mackin (*2*). The presence of *D. marinum* has been demonstrated by this method in oysters from localities in Florida, South Carolina, and Louisiana.

A satisfactory culture medium is Difco's fluid

[1] Grateful acknowledgment is made to Asa C. Chandler for direction of this research, and to Mrs. E. K. Hake for histological assistance. Photomicrographs are by F. C. Breckenridge, Baylor Medical School, Houston.