Fractions 29–44 contained an infusible solid in each flask after the solvent was evaporated. When ether was added the precipitate swelled and became very gelatinous. Following hydrolysis of several of the above cuts with hydrochloric acid, extraction with ether, and evaporation of the solvent, an oily residue remained indicating that the acids had come through the column as aluminum salts. Since a solid residue was recovered from cut 44 after hydrolysis, cuts 29–44 inclusive were combined, hydrolyzed, extracted with ether and dried over sodium sulfate. After the ether was evaporated on a steam-bath, 90 mg. of solid material was separated from the above cuts by crystallization from acetone at $-20°$.

The chromatographic column was next cut into four sections each of which was eluted with methanol. The three lower sections contained gelatinous precipitates which, after hydrolysis, yielded solid residues. One hundred and seventy-five milligrams of solid material (m. p. 50°) was separated from the column and the filtrates. By means of six crystallizations from acetone at $-20°$ in a centrifuge tube, the melting point of this solid was raised from 50 to 69.5°. A mixed melting point with authentic stearic acid was 70° (stearic acid, m. p. 70°). All melting points were taken on a microscope hot stage. The analytical scheme to this point is shown in Fig. 1.

Because the loss of stearic acid during recrystallization was so great, it was necessary to use an indirect method to estimate the amount present. A blank chromatogram containing 3% of stearic acid (42 mg.) in the purified oleic acid was next run. Seventy-four milligrams of solid acid (m. p. 50°) was separated as above. From melting point curves for mixtures of oleic and stearic acids,[8,9] it was established that the isolated material contained 25% of stearic acid, a recovery of 18.5 mg. or 44%.

The solid separated from the bombarded oleic acid (175 mg.) also contained 25% stearic acid (43.8 mg.) as indicated by its melting point of 50°. After correcting for the 44% recovery by chromatography, as indicated above, it was found that the stearic acid isolated (100 mg.) amounted to 5.3% of the non-polymerized saponifiable material. On the basis of the original acid bombarded this figure becomes 1.7%.

Since the stearic acid isolated had been concentrated by a factor of nearly three upon the removal of the polymeric acids and the non-saponifiable material (62.5%), it was necessary to determine if the acid (1.7%) represented

(8) Smith, J. Chem. Soc., 974 (1939).

(9) Markley, "Fatty Acids," Interscience Publishers, New York, N. Y., 1947, p. 124.

original impurity which was below the limit of chromatographic detection using the unbombarded acid. A sample of the purified oleic acid to which was added 1% of stearic acid was, therefore, chromatographed by the above procedure and the stearic acid was easily detectable. No stearic acid could be isolated when the oleic acid itself was chromatographed.

This work indicated that the original oleic acid contained less than 1% of stearic acid and the bombarded oleic acid contained approximately 1.7%, both values being determined by the same analytical technique. It was thus shown that oleic acid was hydrogenated to stearic acid.

Summary

Purified oleic acid has been bombarded with deuterons in the M. I. T. cyclotron. Analysis of the irradiated material has revealed the formation of stearic acid, heptadecene, and polymerized acids.

It has been shown that the hydrogen produced by decomposition of an organic molecule under the influence of radioactivity can enter the double bond of a neighboring molecule. It has thus been demonstrated that hydrogen which may be produced by the effects of radioactivity on the organic constituents of a petroleum source sediment could, in part, be removed from the gas phase by reaction with unsaturated components of the sediments.

(10) Original manuscript received January 28, 1948.

Metal–Salt Interactions at High Temperatures: the Cerium–Cerium Chloride System[1]

BY DANIEL CUBICCIOTTI[2]

The properties of alkali halide crystals containing excess metal have been the subject of extensive investigation by Pohl and co-workers.[3] Their study has led to an explanation of the ability of an alkali halide to dissolve excess metal and to the development of the theory of lattice defect points in crystals.[4] For divalent halides, studies of the miscibility of metal and salt have been

made[5]; however, few other data have been accumulated on such systems. Only one report of metal–salt equilibria for trihalide salts is known to this author. The bismuth–bismuth trichloride and bismuth–bismuth tribromide phase diagrams are reported by Eggink.[6] These systems appear to be similar to those recently reported for divalent halides.[5]

To extend the data in the field of metal–salt equilibria the present study on a trivalent halide was made. The system cerium–cerium chloride was chosen because the melting point of cerium

(1) This work was conducted under the direction of the late Professor E. D. Eastman and sponsored by the Manhattan Project.

(2) Present address: Department of Chemistry, Illinois Institute of Technology, Chicago.

(3) Pohl, Proc. Physical Soc. (London), **49**, extra part, p. 3 (1937).

(4) For a discussion see: Mott and Gurney "Electronic Processes in Ionic Crystals," The Oxford Press, London, 1940.

(5) Cubicciotti and Thurmond, THIS JOURNAL, **71**, 2149 (1949).

(6) Eggink, Z. physik. Chem., **64**, 493 (1908).

is very near that of cerium chloride and thus no elaborate containers were necessary for the materials.

Experimental

Method.—The methods used in this work were the same as those reported in a previous paper.[5] Since cerium metal dissolves iron, however, it was impossible to use iron containers or thermocouple wells. The containers used were molybdenum crucibles formed from sheet metal and welded under a helium atmosphere. The end of the thermocouple well was a molybdenum test-tube of 0.01-inch wall thickness and 4 inches long. This end was machined from a solid rod of metal. It was threaded and screwed into an iron tube to give the thermocouple well sufficient length.

Materials.—Cerium metal and anhydrous cerium trichloride were supplied by Dr. F. H. Spedding of Iowa State College. The trichloride was found to contain 3 weight % other rare earths and less than 0.1 weight % insoluble matter (oxide or oxychloride). The metal had been prepared by the chemical reduction of its chloride and contained 0.4 mole % of chloride. The metal was found to freeze at 770° as compared to a m. p. of 775° given by Kelley.[7] The trichloride froze at 802° as compared to a melting range of 790 to 810° given by Jantsch and Wien.[8]

Results

Some of the cooling curves obtained are shown in Fig. 1 as illustrations of the magnitude of the thermal effects observed. Figure 1, a, b, c, gives curves obtained on the pure salt and salt-rich mixtures. The humped character of the first break indicates some supercooling of the mixture before the solid salt phase separated. The temperatures of the break in such cases was taken as the temperature at the top of the hump.
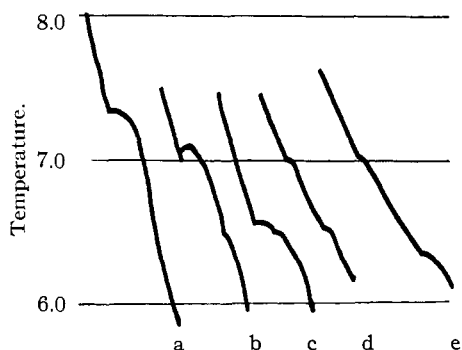

Fig. 1.—Cooling curves of (a) pure cerium chloride, (b) cerium chloride with 10 mole % cerium, (c) cerium chloride with 29 mole % cerium, (d) cerium metal, and (e) cerium metal with 14 mole % cerium chloride (temperature scale in millivolts from a Pt–Pt 10% Rh thermocouple).

In Figure 1, d, e the cooling curves obtained on the metal and a metal-rich mixture are given. In both of these curves there are two breaks detectable. The higher temperature breaks occurred at the same temperature, while the temperatures of the lower breaks are only approximately the same. The higher-temperature breaks for both the metal and the mixture must have been the invariant temperature corresponding to

(7) Kelley, "Contributions to the Data on Theoretical Metallurgy," Bur. Mines Bull. No. 393.

(8) Jantsch and Wien, Monatsh., **69**, 16 (1936).

two liquid plus one solid equilibrium. Apparently, the 0.4 mole per cent. of cerium chloride impurity in the metal was sufficient to lower the freezing point of the metal to the temperature of the three-phase invariant. The lower temperature breaks of these metal-rich mixtures are assumed to correspond to the eutectic break. The temperatures of the lower breaks are below the eutectic break, as observed in the salt-rich region, presumably because the mixture super-cooled.

At one temperature (810°) the compositions of the two liquids in equilibrium were measured. A mixture of salt and metal was equilibrated at that temperature and then quenched by immersing the bottom of the crucible in water.[9] When cold, the metal phase was found on the bottom of the crucible. It had all the appearances of the original metal. The salt-rich phase was an intensely black, friable solid that reacted vigorously with dilute acid.

The temperature–composition diagram for the solid–liquid equilibria of the cerium–cerium chloride system is shown in Fig. 2. Since the cerium metal used was not pure, a literature value for the f. p. of pure cerium is included in the diagram. The point at 775° and 100% cerium is the m. p. of pure cerium as given by Kelley.[7] The shapes of the curves outlining the miscibility gap were assumed to be similar to those observed in other metal–salt systems,[5] and were drawn in through the observed points.
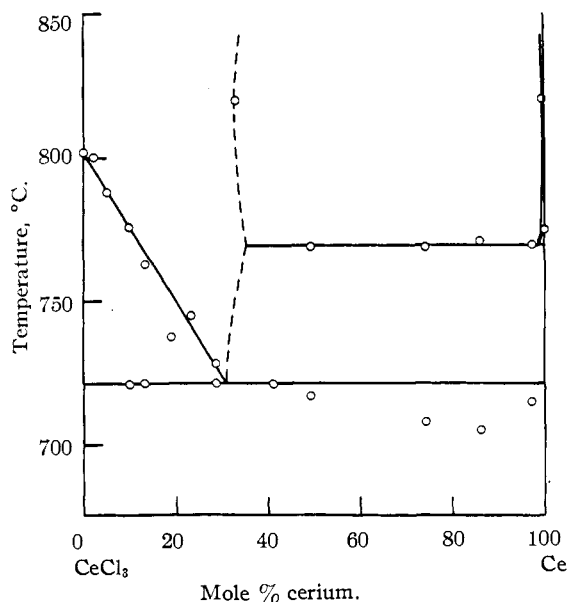

Fig. 2.—The temperature–composition diagram for solid–liquid equilibria in the cerium chloride–cerium system.

Discussion

A comparison of the solubility of cerium in its chloride with the solubilities of other metals in

(9) See ref. (5) for experimental details.

their salts is made in Table I which gives the concentration of metal in the salt phase of the two-phase equilibrium between liquid metal and liquid salt. The metals chosen for the comparison are those whose cationic radii are approximately equal because it has been shown that for divalent systems the solubility depends on the cationic radius.[5]

TABLE I

Metal	Solubility in mole %	Cationic radius[a] in Å.
K	1[b]	1.33
Sr	20[c]	1.13
Ce	33	1.15[d]

[a] Radii from Pauling "The Nature of the Chemical Bond." [b] Estimated from the data of Mollwo, Z. Physik, 85, 56 (1933), on the solubility of potassium in its solid halides and Rogener, Ann. Phys. Leipzig, [5] 29, 386 (1937), on the solubility of potassium in liquid potassium bromide. [c] Estimated from data on other alkaline earth systems, see ref. (5). [d] The radius of trivalent cerium is assumed to be the same as that for trivalent lanthanum.

From Table I it appears that the solubility of a molten metal in its chloride depends upon the charge of the cation of the pure salt. However, this dependence may be a dependence of the solubility upon some property related to the cationic charge. Thus, the significant property may be the ratio of number of cations to number of anions in the salt, which, for a given anion, is proportional to the cationic charge. Additional data would be required to select the property upon which the solubility truly depends.

Summary

The temperature–composition diagram has been determined for the liquid–solid equilibria in the cerium–cerium chloride system. The diagram is similar to those previously reported for some alkaline earth metal–halide systems.

Quantitative Determination of Amino Acids on Filter Paper Chromatograms by Direct Photometry[1]

BY LOUIS B. ROCKLAND AND MAX S. DUNN

The following types of procedures have been suggested for the determination of amino acids on filter paper chromatograms: (A) elution of the spots before or after staining with a chromogenic agent (such as ninhydrin) and analysis of the elutes,[2-8] (B) modified isotope dilution analysis of the spots with the aid of radioactive tracer amino acids,[9] (C) visual comparison of the color intensities of standard and test sample chromatograms stained with ninhydrin,[2] and (D) direct comparison of spot sizes or color intensities of ninhydrin-stained standard and test-sample chromatograms.[10-14] Amino acid test mixtures have been analyzed by Bull, et al.,[11] and Woiwod[3] and protein hydrolyzates by Polson, et al.,[2] Martin and Mittlemann,[4] Keston, et al.,[9] and Block.[13]

(1) Paper 61. For Paper 60, see Murphy and Dunn, Proc. Soc. Exp. Biol. Med., 71, 241 (1949). This work has been aided by a grant from the National Institutes of Health of the United States Public Health Service. The described chromatographic procedures were presented in a symposium before the Division of Chemical Education at the San Francisco, California, meeting of the American Chemical Society, March 30, 1949. The authors are indebted to Jeremiah C. Blatt for technical assistance.

(2) Polson, Mosley and Wyckoff, Science, 105, 603 (1947).

(3) Woiwod, Biochem. J., 42, xxviii (1948).

(4) Martin and Mittlemann, ibid., 43, 23 (1948).

(5) Woiwod, Nature, 161, 169 (1948).

(6) Naftalin, ibid., 161, 763 (1948).

(7) Awapara, Archiv. Biochem., 19, 172 (1948).

(8) Awapara, J. Biol. Chem., 178, 113 (1949).

(9) Keston, Udenfriend and Levy, THIS JOURNAL, 69, 3151 (1947).

(10) Fischer, Parsons and Morrison, Nature, 161, 764 (1948).

(11) Bull, Hahn and Baptist, THIS JOURNAL, 71, 550 (1948).

(12) Block, Science, 108, 608 (1948).

(13) Block, Fed. Proc., 8, 185 (1949).

(14) Fosdick and Blackwell, Science, 109, 313 (1949).

Alanine and glycine have been determined in silk fibroin in the present study by a rapid, direct photometric analysis of color intensities of filter paper chromatograms stained with ninhydrin.

Experimental

Filter paper chromatograms were prepared by the capillary ascent test-tube method of Rockland and Dunn.[15] Ten dilutions, 2×10^{-4} ml. each, of standard containing from 0.625 to 6.250 mg. per ml. of DL-alanine and of glycine were placed on eight replicate strips (10 × 18 × 140 mm.) of Whatman No. 1 filter paper with the aid of a 0.01 ml. total displacement Gilmont ultramicroburet.[16] Similarly, seven dilutions of an acid hydrolysate[17] containing from 3.010 to 12.040 mg. per ml. of silk fibroin (moisture- and ash-free basis) were placed on three replicate strips of the filter paper. The chromatograms were developed simultaneously for three hours at room temperature in 8-inch test tubes containing water saturated phenol, dried for 5 minutes at 100°, sprayed lightly eight times on each side with 0.25% ninhydrin solution in water saturated butanol and heated for five minutes at 100°. The alanine was resolved completely at the seven levels but glycine only at the two lowest levels.

In order to determine the concentrations of the amino acids, the filter paper strips were placed in a special sample holder containing an opening of a size just sufficient to enclose the entire area of the colored spot and the color intensities were read directly with the aid of a photoelectric colorimeter.[19] The alanine and glycine content of silk fibroin were estimated by interpolation from the standard curves drawn from plots of per cent. transmission against concentration of amino acid on coördinate paper.

(15) Rockland and Dunn, ibid., 109, 539 (1949).

(16) Emil Greiner Company, 161 6th Avenue, New York City.

(17) Prepared by Dr. M. N. Camien from the silk fibroin described by Dunn, et al.[18]

(18) Dunn, Camien, Rockland, Shankman and Goldberg, J. Biol. Chem., 155, 591 (1944).

(19) Lumetron, Model 402 EF, manufactured by the Photovolt Corporation, 95 Madison Avenue, New York City.