

## CHEMISTRY

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### Abstracts

**Determination of Phenol Equivalence by Conductometric Titration.** R. LEWIS VAN ATTA and ROBERT E. VAN ATTA, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——Certain household disinfectants, such as Lysol, contain various substituted phenols as active ingredients. The “phenol equivalence” of these solutions may be determined conveniently by conductometric titration with concentrated lithium hydroxide, similarly standardized against liquefied phenol. Titration of at least 300 ml of diluted disinfectant or standard solution with 0.5-ml increments of 2.5 *N* titrant, using a conductivity dip cell ( $K = 1.0$ ) and a conventional conductance bridge to monitor changes in solution conductance, yields titration curves permitting rapid, accurate location of end-points at the intersection of straight lines.

**Gas Chromatographic Determination of Methyl Salicylate in Rubbing Alcohol by Standard Addition.** ROBERT E. VAN ATTA and R. LEWIS VAN ATTA, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——Certain commercial rubbing alcohol preparations contain 1-3 volume per cent of oil of wintergreen, added due to its pleasant odor and penetrating properties. These preparations may be conveniently analyzed for methyl salicylate content by gas chromatography with a carbowax column at oven temperatures of 175-200°C. The ester peak is symmetrical and cleanly resolved, appearing well after elution of the major components isopropyl alcohol and water. The peak height, measured from the extrapolated background base-line, is proportional to concentration. The analysis is rapid and accurate, either by graphical evaluation or appropriate calculation from unknown and standard addition peak heights.

**Spectrophotometric Determination of Caffeine in Cola Drinks.** ROBERT E. VAN ATTA, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——Caffeine may be quantitatively determined in 5-ml aliquots of cola-containing soft drinks, made basic with 20%  $\text{Na}_2\text{CO}_3$ . The caffeine is extracted with two 20-ml portions of dichloromethane and measured spectrophotometrically at 277 nm,  $\lambda_{\text{max}}$  for the ultraviolet absorption of caffeine in that solvent. Identical extractions and analyses of suitable volumes of 0.40 mg per ml standard caffeine solution yield a linear calibration curve up to 2 mg of caffeine per ml. Benzoic acid, present in the aqueous layer if the soft drink contains sodium benzoate, is not extracted with the caffeine.

**Turbidimetric Determination of Lead in Pottery Extract.** ROBERT E. VAN ATTA, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——Lead, extracted from pottery by 4% acetic acid, may be quickly and accurately determined by precipitation as lead chromate. The precipitate is formed in a medium containing a glycerol-ethanol mixture as conditioning agent. The analysis is completed by turbidimetric measurement of the suspension, contained in 18 x 150 mm glass cuvettes, at 510 nm in a Spectronic 20 spectrophotometer or the equivalent. The composition of the extract is determined by reference to an appropriate calibration plot, apparent %T vs.  $\mu\text{g}$ . Pb per ml. If the suspension is carefully prepared, mixed, and timed prior to measurement, the calibration plot is linear up to 15  $\mu\text{g}$  per ml.

**Chemical Analysis of the Quality of Surface Water of the Wabash River at Terre Haute, Indiana.** JOSEPH R. SIEFKER and LAWRENCE W. FRENCH, Department of Chemistry, Indiana State University, Terre Haute, Indiana 47809.——The purpose of this study was to record certain qualitative chemical characteristics of water samples taken from the Wabash River, and to determine quantitatively the amounts of calcium, chloride, dissolved oxygen, dissolved solids, fluoride, iron, lead, magnesium, manganese, mercury, potassium, sodium, total solids, turbidity, and zinc in the samples. Quantitative chemical data were determined for more than 4,500 separate analyses of approximately 175 water samples. Water samples were collected over a one-year period, one to six times per month, at seven sites, including a site at Coal Creek and the Terre Haute Wastewater Treatment Plant, and five sites along the Wabash River.

**Reaction of Silicic Acid with Fluoride.** EUGENE SCHWARTZ, Chemistry Department, DePauw University, Greencastle, Indiana 46135; R. H. BUSEY and R. E. MESMER, Chemistry Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee 37830.——Potentiometric titrations were used to determine the products formed when fluoride was reacted with monomeric silicic acid,  $\text{Si}(\text{OH})_4$ , in acid solutions. The free fluoride ion concentration was monitored with a fluoride electrode and the pH with a quinhydrone electrode. Measurements were made in both perchlorate and chloride media at 0°, 25°, and 60° C. Data were analyzed and results are reported in terms of  $\bar{n}(\text{H})$  and  $\bar{n}(\text{F})$ , the average number of hydrogen ions and of fluoride ions reacted per silicic acid molecule. The predominant product is  $\text{SiF}_6^{2-}$ . The equilibrium constant for the reaction

$$\text{Si}(\text{OH})_4 + 4\text{H}^+ + 6\text{F}^- = \text{SiF}_6^{2-} + 4\text{H}_2\text{O}$$

is about  $10^{30}$  at 25° C in a chloride medium. The behavior of silicic acid toward fluoride is in contrast to that of boric acid where the formation of the fully-fluoridated product  $\text{BF}_4^-$  from the precursor  $\text{BF}_3(\text{OH})^-$  is kinetically hindered.

**Substitution Reactions of Tungsten(O) Complexes.** TROY L. BALLARD, JOHN A. MOSBO, and BRUCE N. STORHOFF, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——Reactions of  $\text{W}(\text{CO})_4(\text{TMPA})$  (TMPA = N, N, N', N'-tetramethyl-1,3-diaminopropane) with monodentate phosphine and phosphite ligands with cone angles between 105° and 180° have been investigated. The conditions required to completely displace the chelating diamine have been determined and spectroscopic procedures developed to determine the relative amounts of *cis* and *trans* isomers formed. For example, the reaction of a large excess of diphenylmethoxy-phosphine or

diphenylmethylphosphine (cone angles of  $136^\circ$  and  $132^\circ$ , respectively) with  $W(CO)_4(TMPA)$  yields *cis* complexes.

The potential for using the *cis-trans* ratio of the products as an indicator of ligand size will be discussed.

**Studies of 7-Hydroxychromones.** GERALDINE M. HUITINK, Department of Chemistry, Indiana University at South Bend, South Bend, Indiana 46615.——A series of 7-hydroxychromones substituted at positions -2 and -3 with methyl and phenyl groups has been studied. One member of the series, 7-hydroxy-2,3-dimethylchromone, exhibits properties that make it suitable for use as the fluorescent moiety in metallofluorescent indicators.

**Chelation of Dialcohols with the NMR Shift Reagent  $Eu(fod)_3$ .** T. L. KRUGER, J. A. MOSBO and R. PRATT, Ball State University, Muncie, Indiana 47306.——The interactions of difunctional molecules with NMR shift reagents has been studied using dialcohols and tris (1,1,1,2,2,3,3-heptafluoro-7,7-dimethyl-4,6-octanedione) europium(III) ( $Eu(fod)_3$ ). Equilibrium constants of complex formation were obtained from plots of chemical shifts vs. ratios of shift reagent to substrate concentrations. Chelation vs. monodentate coordination was determined from these data and from chemical shift changes induced by competition from a monofunctional alcohol. The effects of chelate ring size, conformational changes and steric interactions upon chelate formation were observed.

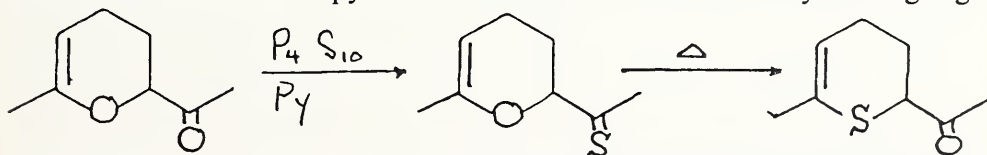
**Non-Metal Pi-Face Bonding of Cyclobutadiene.** K. B. LIPKOWITZ, Department of Chemistry, Indiana University-Purdue University at Indianapolis, 1201 East 38th Street, Indianapolis, Indiana 46227.——Because the gas phase lifetime of cyclobutadiene is on the order of milliseconds, it is not amenable to routine spectroscopic investigation. Generation of cyclobutadiene in noble gas matrices at very low temperature extends its lifetime so that spectroscopic studies can be accomplished. Typically an appropriate precursor is trapped in the matrix and photolyzed. Cyclobutadiene is generated but so are several photo by-products. A theoretical investigation of the pi-face bonding of cyclobutadiene with these by-products is presented.

**Studies of Oxygen Exchange Kinetics Using  $^{31}P$  and  $^{13}C$  Nuclear Magnetic Resonance Spectroscopy.** JOHN M. RISLEY and ROBERT L. VANETTEN, Department of Chemistry, Purdue University, West Lafayette, Indiana 47907.——The study of the kinetics of oxygen exchange reactions has usually involved a discontinuous assay of the system of interest; the procedure requires stopping the reaction at time intervals, isolating the compound(s) of interest, in some cases derivatizing the sample, and finally analyzing the derivative mass by spectroscopy. Cohn and Hu [Cohn, M. and Hu, A. (1978) *Proc. Natl. Acad. Sci. USA* 75:200-203] recently described the  $^{18}O$ -isotope effect on the resonance signal of the  $^{31}P$  nmr spectrum of inorganic phosphate. The  $^{18}O$ -isotope effect on the  $^{31}P$  nmr signal of inorganic phosphate is a 0.021 ppm upfield shift per  $^{18}O$  atom bonded to phosphorus. The  $^{18}O$  induced shift on  $^{31}P$  nmr spectra has already been used in studies of enzyme-catalyzed exchange reactions involving phosphate-phosphate and phosphate(oxygen)-water systems [Bock, J. L. and Cohn, M. (1978) *J. Biol. Chem.* 253:4082-4085 and Webb, M. R., McDonald, G.

G. and Trentham, D. R. (1978) *J. Biol. Chem.* **253**:2908-2911]. The phosphate (oxygen)-water exchange catalyzed by homogeneous human prostatic acid phosphatase has been studied in our laboratory using highly enriched  $^{18}\text{O}$ -phosphate [VanEtten, R. L. and Risley, J. M. (1978) *Proc. Natl. Acad. Sci. USA* **75**:4784-4787].

A comparable but potentially even more widely applicable observation would be an  $^{18}\text{O}$ -isotope effect on the resonance position of carbon-13 in  $^{13}\text{C}$  nmr spectroscopy. Jameson [Jameson, C. J. (1977) *J. Chem. Phys.* **66**:4983-4988] predicted such an upfield shift in the  $^{13}\text{C}$  nmr spectrum of  $\text{CO}_2$  and it should depend on the number of  $^{18}\text{O}$  atoms bonded to the carbon atom. We have now observed such an upfield shift in the  $^{13}\text{C}$  nmr resonance signal of both an alcohol and a carboxylic acid; the  $^{18}\text{O}$ -isotope effect is 0.035 ppm per  $^{18}\text{O}$  atom. These results are of particular utility in studies of carbon-oxygen exchange kinetics. We have demonstrated the value of the  $^{13}\text{C}$  nmr shift induced by  $^{18}\text{O}$  by measuring the rate of loss of the  $^{18}\text{O}$  label from a sample of  $^{18}\text{O}$ -t-butanol; the resultant kinetic data agrees with the literature value obtained by conventional mass spectrometry [Dostrovsky, I. and Klein, F. S. (1955) *J. Chem. Soc.* **1955**:791-796]. Thus the  $^{18}\text{O}$ -isotope induced shifts of  $^{31}\text{P}$  and  $^{13}\text{C}$  nmr signals provide valuable tools for the study of kinetics, particularly since they provide continuous, non-destructive assays.

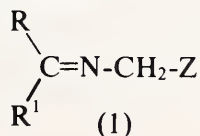
**Formation and Synthetic Utility of Dihydropyrans.** K. B. LIPKOWITZ and S. SCARPONE, Department of Chemistry, Indiana University-Purdue University at Indianapolis, 1201 East 38th Street, Indianapolis, Indiana 46227.\_\_\_\_\_Several 2,3-dihydropyrans have been prepared via the thermally allowed 4+2 cycloaddition of  $\alpha,\beta$ -unsaturated carbonyls. Since these rings are 1,5-dienes they can and do rearrange via the symmetry allowed [3,3] sigmatropic shift. Functionalization of the pyrans can then lead to new heterocyclic rings eg:



The synthesis of thiapyrans and attempted syntheses of spirocyclic natural products using this methodology is presented.

**The Synthesis of Amino Acids by Phase-Transfer Reactions.** MARTIN J. O'DONNELL, THOMAS M. ECKRICH, ROBIN L. POLT, GILBERT S. STATEN and KATHLEEN R. VOGLER, Department of Chemistry, Indiana University-Purdue University at Indianapolis, 1201 East 38th Street, Indianapolis, Indiana 46205.\_\_\_\_\_A simple synthetic route is presented for the preparation of higher amino acids from the simplest amino acid, glycine. A key step in this sequence is the novel use of phase-transfer alkylations of glycine derivatives.

Various Schiff base derivatives (1) of glycine ( $\text{R} = \text{aryl}$ ;  $\text{R}^1 = \text{aryl}$ ,  $\text{H}$ ;  $\text{Z} = \text{CO}_2\text{R}^{11}$ ,  $\text{CN}$ ;  $\text{R}^{11} = \text{alkyl}$ ) are readily prepared by the condensation of a ketone or aldehyde with the appropriate amine.



Compounds *I* can be alkylated in good yields by phase-transfer alkylations. The alkylated derivatives are then readily hydrolyzed to the higher amino acids. The effect of variation in the structure of *I* and the reaction conditions on the outcome of the phase-transfer reactions will be discussed.

**The Isolation of the Two Dominating Forms of Adenosine Deaminase from Frog Tissues.** KENNETH S. COOK and PANG FAI MA, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——Studies of adenosine deaminase (adenosine aminohydrolase, EC 3.5.4.4) from mammals, reptiles, amphibians, and avians have shown three molecular forms of 200,000, 100,000, and 35,000 daltons, designated as A, B, and C forms, respectively. Ma and Fisher (1968) have shown that the tissue specific differences, with respect to adenosine deaminase in the chicken, were due to the presence of a B form enzyme in the liver which was absent in the duodenum. A C form enzyme was present in both tissues and has shown strikingly similar properties. Tissue specific differences have been reported when various frog tissues were compared. This study will attempt to isolate the two enzyme forms from frog tissue and determine a few kinetic parameters, such as the relative substrate specificities, the Michaelis constant, and the energies of activation. The results will serve as a basis to explain the differences observed in different tissues. A procedure for partial purification of the enzyme forms will also be discussed.

**Effects of Ionic Strength and Buffer Composition on the Heat Stability of Bovine Erythrocyte Superoxide Dismutase.** DOUGLAS B. WILLIAMS and ERIC R. JOHNSON, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——Copper-zinc superoxide dismutase (E.C. 1.15.1.1) isolated from bovine erythrocytes has been found to exhibit an unusual resistance against irreversible denaturation at temperatures of 80-100°C. This heat stability is markedly dependent on ionic strength, with the enzyme showing increasing stability with decreasing ionic strength. The greatest heat stability was observed with a superoxide dismutase solution that had been exhaustively dialyzed against distilled water. The polyvalent anions, sulfate and phosphate, were found to decrease the heat stability of the enzyme to a greater extent than the monovalent chloride anion.

**A Positive Ribonuclease Zymogram Technique.** EMILY A. YOUNT, ROBERT C. KARN, MARGARET L. CRISP, and M. E. HODES, Department of Medical Genetics, Indiana University, Indianapolis, Indiana 46202.——A zymogram technique is presented in which the presence of ribonuclease is demonstrated by a dark band on a light background. This system has several advantages over previous methods in which the presence of ribonuclease is demonstrated by a light cleared area on a dark background of stained RNA (negative staining methods). This new technique is more sensitive, gives better resolution, takes less staining time, and is less subject to interference by nonenzymatically active protein than previous methods. This new technique involves digestion of dinucleoside monophosphate substrates of the type Pyr-p-A to produce adenosine. A series of enzymatic reactions is used to link adenosine production to reduction of a tetrazolium salt and darkening of the gel. We are presently using this technique to screen for polymorphisms in ribonucleases from human lymphocytes and granulocytes.