either in the columns of the JOURNAL or to him personally. He is earnestly desirous of knowing what is in the minds of the Conference membership. A definite plan must be presented for action at the next meeting of the Conference at New Orleans. This is necessary in order that Conference membership may mean something and in order that the Conference may exert a greater influence in matters educational and legislative.

(Signed) R. A. LYMAN,
Chairman Executive Committee,
American Conference of Pharmaceutical Faculties.

THE APPLICATION OF OPTICAL METHODS TO THE EXAMINATION OF INSECTICIDES AND FUNGICIDES.*

BY GEO. L. KEENAN.

In the course of the microscopical examination of products for the Insecticide and Fungicide Board, the writer has found it advantageous to make use of optical (and microchemical) tests for the identification of the ingredients contained in the various samples submitted. The application of optical-crystallographic measurements has heretofore been largely employed for the identification of minerals, especially as they occur in rocks or as they are produced synthetically. Recently Dr. Edgar T. Wherry, Crystallographer of the Bureau of Chemistry, has extensively used optical-crystallographic methods in the identification of alkaloids and other organic compounds. Such determinations have been found to be of considerable assistance to the chemist in the identification of these products. The success attending the use of such data led the writer to consider their application to the identification of ingredients commonly found in insecticides and fungicides.

A large number of the insecticides and fungicides submitted for microscopical examination are simple mixtures, consisting of some powdered vegetable materials, with or without varying proportions of inorganic and organic substances, or consisting wholly of inorganic materials. The product is often of such a nature that it can be readily separated into portions by sifting through a 40-mesh or 60-mesh sieve. The very fine powder which passes through the sieve is subjected to close scrutiny under the microscope. The coarser portions remaining upon the sieve are spread upon a sheet of paper and examined with the aid of a hand lens (magnifying about 10 diameters). Various separations can be made with the aid of forceps and particles removed which can be examined as to their optical properties.

For the purpose of studying the optical properties of any crystalline fragments, it is essential that the microanalyst be provided with a microscope equipped with nicol prisms. An ocular with cross-hairs, a 4-millimeter, an 8-millimeter, and a 16-millimeter objective are usually all the necessary lenses that are required. A slit should be provided in the microscope tube above the objective and below the upper nicol prism for the insertion of a selenite plate, commonly designated as "red, 1st order." A rotating stage, an Abbé condenser, and a substage iris

* Contribution from Insecticide and Fungicide Board and Bureau of Chemistry, Department of Agriculture, Washington, D. C.

diaphragm are also quite necessary. Liquids of known refractive index, usually oily in character, are needed, in which to immerse the crystal fragments. The value of the index of each liquid is first determined by means of a refractometer. For most identification purposes, the liquids used differ in index only by two units in the second decimal place, that is, 1.48, 1.50, 1.52, 1.54.

Fragments of crystalline material picked out of a sample are crushed on a microscopical slide and immersed in any one of the liquids whose refractive index has already been determined on the refractometer. Observations are made according to the following procedure:

I. EXAMINATION IN ORDINARY LIGHT.

(*a*) The color of the fragments in ordinary light is noted. Occasionally the general appearance of the substance may furnish some clue as to its identity.

(*b*) The general outline or shape, technically described as the crystal habit, is also recorded.

II. EXAMINATION IN PARALLEL POLARIZED LIGHT, NICOLS CROSSED.

(*Parallel polarized light is produced by inserting the crossed nicol prisms.*)

(*a*) The nature of the extinction of the material, whether parallel or inclined, is determined. This is arrived at by placing the long axis of a crystal parallel to one of the cross-hairs of the ocular. The angular reading on the stage is recorded. Then the crystal is turned to the point of maximum darkness. The reading on the stage is again recorded. If there is any difference between the two readings, the crystal has inclined extinction; otherwise parallel extinction. Several crystals are usually examined and an average of the results taken. In the tetragonal, trigonal, hexagonal and orthorhombic systems the extinction is normally parallel, as it is also, in some instances, in the monoclinic system. Crystals of the monoclinic and triclinic systems, however, usually show inclined extinction. Crystals belonging to the cubic system do not change from light to dark in parallel polarized light, but remain dark in all orientations.

(*b*) The sign of elongation is commonly determined as follows: A long and narrow crystal, showing very little color with nicols crossed, is so oriented that its long dimension is parallel to direction "*c*" of the selenite plate which is inserted in the slit provided for it in the microscope tube. (Direction "*c*" of the selenite is commonly indicated by an arrow engraved on the plate.) If the crystal appears blue (or green) in color, the elongation is positive; if the crystal appears yellow, white or gray, the elongation is negative.

III. EXAMINATION IN CONVERGENT POLARIZED LIGHT, NICOLS CROSSED.

(*Convergent polarized light is produced by bringing the substage condenser as close to the slide as possible and inserting the 4-millimeter objective.*)

(*a*) In order to determine the crystal system to which any crystalline fragment belongs, it is essential to determine the character of the extinction, whether parallel or oblique, and also to obtain some indications of what is known as an interference figure. In doubly refractive substances (*i. e.*, those which become alternately light and dark when the stage is rotated with nicols crossed), the interference figure shown may be uniaxial or biaxial. To obtain the best figures,

fragments of material should be chosen which remain dark or at most faintly illuminated during complete rotation of the stage. After locating and centering the grain to be examined, the ocular is removed and the nature of interference figure observed by looking through the microscope tube. In the case of a uniaxial crystal, a dark cross appears in the field, surrounded by rings, the figure remaining more or less stationary when the stage is rotated. A biaxial interference figure consists, in the ideal case, of two dark spots, surrounded by concentric rings, each dark spot being crossed by a curved black bar. If a uniaxial figure is shown and the extinction is parallel the indications are that the fragment belongs to either the tetragonal, hexagonal or trigonal system. A biaxial figure on a substance with parallel extinction, indicates the orthorhombic system, with inclined extinction either the monoclinic or triclinic system. (Crystals elongated in the direction of the *b*-axis show parallel extinction in the monoclinic system.)

(*b*) The optic sign is indicated as being positive or negative, as the case may be. It is determined by inserting the selenite plate and noting the change of color on opposite sides of the central cross of the interference figure. If areas of blue occur in a direction parallel to *c* of the selenite plate the sign is positive. If the color is yellow in that direction, the sign is negative.

IV. DETERMINATIONS OF THE REFRACTIVE INDICES OF THE SUBSTANCE.

In many instances the crystal system to which the fragment belongs may not be easily determinable, due to the difficulty encountered in obtaining good interference figures and also in positively determining the character of the extinction exhibited. Notwithstanding, other constants can be determined which are of value in proving the identity of the substance. Fortunately for the microanalyst, a great majority of the commonly occurring crystallizable inorganic chemical salts have been studied optically and at least sufficient data obtained on them to make their identification sure.

The most important optical constants which serve to identify a product with certainty are the refractive indices. These vary in number, depending upon the system in which the substance crystallizes. In crystals of the triclinic, monoclinic, and orthorhombic systems, three indices can be determined, the smallest being designated α, the intermediate β, and the largest γ. In those belonging to the hexagonal, tetragonal, and trigonal systems, two indices can be determined, ω and ϵ. Crystals belonging to the cubic system give but one index, indicated as n.

The fragment of material is immersed in the liquid on a slide, and examined under the microscope with one nicol prism in place; attention is then paid to the direction in which a band of light, which will be seen surrounding the fragment, moves when the tube of the microscope is raised. If the band of light passes into the crystal, it indicates that the refractive index in that crystallographic direction which chances to lie parallel to the plane of the nicol is higher than that of the liquid; if it passes out of the crystal the index of the liquid is higher than that of the crystal in that crystallographic direction. In other words, when the microscope tube is raised, the band of light passes toward that medium (either liquid or crystal) which has the higher index of refraction. When the crystal

(in any particular position) and the immersion liquid are of the same refractive index, the crystal fragment disappears completely in the liquid. Therefore the process is repeated with different liquids until the crystal completely matches the liquid, that is, no band of light passes either into or out of the crystal. A large number of fragments are examined until the analyst is convinced that the minimum, intermediate, and maximum indices in the case of the monoclinic, triclinic, and rhombic systems, or two indices in the case of the hexagonal, tetragonal, and trigonal systems, have been determined. Only one index can be determined for the cubic system. Two indices can be observed on any one fragment (if it has more than one), the index crosswise of an elongated crystal being shown when this crystal is perpendicular to the direction of vibration of the nicol, and the index lengthwise when the crystal is parallel to this direction.

Monochromatic Light.—The refractive index of a substance varies with the wave length of the light. It is customary to use the index for the *D*-line (sodium light) as the typical one for ordinary substances. For some purposes the average index, obtained by using white light, is sufficient. However, with some substances the band of light used for determining refractive indices is more or less separated into colored bands when white light is used, rendering the phenomenon difficult to observe. In general, therefore, the microanalyst should avail himself of some form of monochromatic light. This may be produced by using the light from a sodium flame, or by passing white light through some form of ray-filter. Wherry has recommended the use of a combination of Wratten *E*-red No. 23 and *B*-2 Extra Light No. 57-*A* filters, 3 inches square, in glass. It may be noted that the light transmitted by the yellow glass commonly used in automobile lamps is sufficiently monochromatic for most purposes.

EXAMPLE.

A sample was sifted through a 60-mesh sieve and the portion remaining on the sieve spread out upon a sheet of white paper for examination with a lens. Among the various fragments noted were small particles of a bluish color. One of these was removed to a slide, crushed, and a small drop of liquid added, the product then being examined in ordinary light.

I. EXAMINATION IN ORDINARY LIGHT.

The fragments were colorless and irregularly angular.

II. EXAMINATION IN PARALLEL POLARIZED LIGHT, NICOLS CROSSED.

(*a*) In parallel polarized light the colors exhibited by the majority of the particles were very striking. Some of the particles were more or less colorless.

(*b*) The particles were so irregular in shape that it was difficult to determine the extinction angle. Some particles more or less rectangular in shape indicated that the extinction was inclined.

(*c*) The sign of elongation could not be satisfactorily determined because of the nature of the material.

III. EXAMINATION IN CONVERGENT POLARIZED LIGHT, NICOLS CROSSED.

(*a*) One of the larger colorless fragments was located with the low power objective. The 4-millimeter objective was inserted and the substage condenser

brought into place. The particle was carefully centered. The eyepiece was removed and the stage slowly rotated. As the stage was rotated a distinct curved black bar passed into the field of vision and soon disappeared. The concentric rings already referred to were not very distinct but the curved bar was clearly a portion of a biaxial interference figure. The character of the extinction and the interference figure obtained indicated that the material belonged to either the monoclinic or triclinic system.

(*b*) Interference figure showing concentric rings was not observed, therefore the optic sign could not be determined with certainty.

IV. DETERMINATION OF THE REFRACTIVE INDICES.

A crushed fragment was immersed by way of preliminary trial in a liquid with a refractive index of 1.65. With the lower nicol in position, the band of light surrounding a large number of particles was found to pass from the crystal toward the liquid when the microscope tube was raised. This phenomenon was exhibited when the fragment was perpendicular to the vibration direction of the nicol as well as in the other position. It was apparent that the refractive index of the liquid was considerably higher than that of the crystal.

More material was immersed in liquid with index of 1.49. On raising the microscope tube, the band of light surrounding the crystal fragments passed into the crystals. This indicated that the index of refraction of the crystals was higher than that of the liquid, this being the case in every crystallographic direction noted.

Another fragment of the material was crushed and mounted in cedarwood oil which had an index of 1.515. Rotating the stage of the microscope then caused a large number of the irregular fragments to completely disappear. Examination of a large number of fragments indicated that no index was lower than 1.515.

Other fragments of the material showed an index appreciably higher than 1.515. The substance was immersed in liquid 1.540; many of the particles exactly matched this liquid. No index higher than 1.540 seemed to be present in the material. Evidently the minimum and maximum indices for the substance had been determined to be 1.515 and 1.540, respectively.

An intermediate index was indicated on irregular fragments showing very little color when the nicols were crossed. These particles when immersed in practically anhydrous clove oil with an index of 1.535, disappeared (as far as could be determined in monochromatic light). Confirmation of this fact by the examination of a large number of fragments determined the intermediate index to be 1.535.

Summarizing the data obtained, the following conclusions could be drawn concerning the identity of the substance in question. The biaxial interference figure and the inclined extinction indicated on the fragments suggested that the substance crystallized in either the monoclinic or triclinic system. This being the case, the three indices obtained could be indicated as follows: α: 1.515, β: 1.535, γ: 1.540. Reference to a table containing optical data of many of the commonly occurring crystallizable salts identified the substance as being copper sulphate, $CuSO_4.5H_2O$. This conclusion was further substantiated by studying some known material and comparing the results obtained with those character-

istic of the substance under examination. (In many instances microchemical tests are also of value for the purpose of verification.)

SUMMARY.

Attention has been called to the application of optical-crystallographic methods for the identification of crystallizable chemical salts in insecticides and fungicides. The author has found the use of such methods of value in many instances and has attempted to outline a simple method of procedure particularly suitable for microanalysts who have not had extensive training in crystallography and mineralogy.

No attempt has been made to go into detail concerning crystallographic and optical concepts, it being left to the reader to avail himself of these in works dealing primarily with petrographic methods.

THE DELICACY OF THE U. S. P. TESTS.

BY JOSEPH ROSIN.

By "delicacy" is here understood the quantity or proportion of the impurity examined for which will just fail of being detected. It is practically synonymous with "sensitiveness" and the two terms are used here interchangeably.

It is almost needless to state that in making the U. S. P. tests the directions of the Pharmacopoeia were closely followed. In several instances, however, deviations from the Pharmacopoeial instructions were necessary, and these are noted under the respective tests. Where the U. S. P. directions are not sufficiently explicit, the more explicit details given in the U. S. P. for testing other similar products for the same impurities were observed, or the established good practice in carrying out tests of this character was followed.

The materials used were the purest obtainable, or they were specially prepared. If they were not entirely free of the impurities to be tested for, the latter were determined and a correction applied in the final calculations of the results.

The method of procedure was the one generally employed in an investigation of this character. Solutions or mixtures of the pure material with varying proportions of the impurity to be tested for were subjected to the U. S. P. test or to the proposed test.

Before proceeding with the presentation of the results it will be well to note that in instances where the U. S. P. allows a slight turbidity or opalescence in the test, the results recorded are necessarily arbitrary, because, as it has been pointed out by others, the meaning of "slightly turbid" or "opalescent" is subject to various interpretations by observers. Even in the more definite tests where no manifestation of the presence of the impurity is allowed, somewhat divergent and occasionally very divergent results are obtained, because of differences in the manipulation of the test. For instance, in the majority of the U. S. P. tests it is simply directed to add the reagent to the solution or substance under examination without stating whether it should be mixed. True, the accepted practice is to mix the solution, but it is also true that not all mixings are equally effective. This is particularly the case when the quantities tested for are small and the time limit for the reaction is short. Other details in the manipulation, the diameter