not the most serious, for it can be taken care of by correspondingly increased wages. If the cost of living, measured in dollars, were doubled, and wages also were doubled, we should be as well off as before. Real hardship comes only if prices advance at a more rapid rate than wages. Whether this takes place or not depends—aside from unjust operations of monopolists and profiteers—very largely on the productive efficiency of our industries. And we may well doubt whether any one single agency equals, in its latent power to multiply this efficiency, the almost untapped reserve of coöperation in scientific and technical research.—*Scientific American.*

THE DEVELOPMENT OF QUANTITATIVE MICROSCOPY.

BY T. E. WALLIS,* B.SC. (LOND.), F.I.C.

Quantitative microscopical methods are being slowly developed by a small number of isolated workers both in Europe and in America and a certain amount of substantial progress has been made. The subject is now so far advanced that a general review of the situation seems desirable and a free discussion of methods would help those concerned to build upon a sure foundation.

The whole subject is surrounded by so many difficulties that its advocates find their progress hindered by a lack of reliable data upon which to proceed. These fundamental factors are slowly accumulating and can be accepted only when based upon very careful investigations.

The two subjects needing closest attention appear to be (*a*) the limitation of effort to work upon material that promises to yield satisfactory results, and (*b*) the elaboration of a universally applicable method of procedure.

SELECTION OF SUITABLE MATERIAL.

The materials suited for accurate estimation by microscopical counts are limited, at present, to such as contain naturally formed particles of small size such as pollen-grains, starch grains, spores, etc. Such particles are recognizable as intrinsic units that are readily identified by all workers and, when counts are made, they will be satisfied that each one has been working along the same lines.

It is, therefore, to substances containing such particles that one's energies are best directed and it would seem wise to concentrate upon these before attacking the more difficult problems which will involve additional measurements, such as length and area.

In the present state of our knowledge, substances such as sand, powdered sulphur, charcoal and sugar, where the number of particles per milligramme will obviously vary enormously according to the degree of comminution, cannot be successfully estimated by counting methods and it would seem wisest to avoid expenditure of efforts upon such determinations. In the great majority of instances —if not in all—substances such as these can be accurately estimated by chemical and physical methods and this being so, microscopical methods become superfluous. To attempt to make such determinations microscopically tends to bring discredit upon microscopical methods generally.

The counting of fibers and stone cells in a way that will stand severe critical examination and find acceptance among analysts generally is an extremely diffi-

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cult problem and hitherto no satisfactory suggestion for surmounting it has been made. One can hardly expect the critical observer to accept counts of masses of fibers of uncertain length and in bundles of varying numbers, as has been suggested by some workers.[15] Before an acceptable process can be devised, it will be necessary to discover some method of eliminating the uncertainties resulting from these disturbing factors.

Even when dealing with such apparently simply constituted materials as starch, many difficulties arise and they are of such importance that one must regard figures giving the number of starch grains per milligramme of various starches as only roughly approximate. That this is so, is evident from a consideration of the results given by different investigators.[8, 14, 17, 18]

One cause of this variation is due to the difficulty of deciding exactly what is a typical grain of starch. For example, some workers stain with iodine and count all stained particles and, if this same starch were counted without staining, there are many minute specks which would be passed over as not conforming to the appearance of typical grains. It seems preferable to count starch unstained and only to include such grains as can be definitely recognized as starch.

Another source of variation will be found in the enormous difference produced by different methods of preparation. If a starch, like wheat, is washed by sedimentation for short periods, the resulting starch will show many less grains per milligramme than if the washings are less numerous and the periods of sedimentation longer. This is because many more of the smaller grains are retained by the second process than by the first. The same remark applies in a varying degree to all starches and until a standard method of preparation of the starch is adopted, no concordance of results can be expected.

It seems desirable in the first place to secure concordant results by different workers for a substance which is much less variable than starch. Probably the most uniformly constituted commercial vegetable substance is lycopodium and this material seems, therefore, to be the most suitable from which to make a start in quantitative microscopy. The number of spores per milligramme has been determined and found to be 94,000.[17] One would like to see this figure investigated by others and to accumulate results from workers in different places, so that absolute confidence may be established in some fundamental figure as a starting point for all similar work.

METHOD FOR MAKING COUNTS.

A method of counting that will be universally acceptable among analysts must be one that can be applied under any ordinary conditions and with an ordinary microscopical outfit. Special stages, mechanical appliances and slides must, if possible, be eliminated and the mixings must be rendered as simple as possible. The method must be independent of the magnification used and the particular lenses employed to produce the magnification; it should be simple in principle and suited for use with a very wide range of substances.

Of the various methods proposed, the Lycopodium Method introduced by the author[16, 18] is the only one which possesses all these advantages. It is extremely simple in principle and easy to carry out in practice. Briefly stated, the method consists in mixing known weights of lycopodium and of the material to be

examined and suspending the mixture in any convenient volume (not necessarily a known volume) of a suitable suspending agent such as mucilage of tragacanth, olive oil, castor oil or a mixture of these oils. (For a discussion of suspending agents see reference 16.) The suspension is thoroughly shaken up in a stoppered weighing bottle or in a corked tube and a small drop is transferred to a slide by means of a glass rod and covered with a cover-glass.

Counts are made in a number of fields, whose positions are selected beforehand so as to use some from all parts of the preparation, of both lycopodium spores and characteristic elements of the powder to be analyzed. Since there are 94,000 spores per milligramme of lycopodium, one can immediately calculate how many characteristic particles are present for every milligramme of lycopodium, and from the known composition of the mixture of spores and substance to be examined one immediately finds the number of characteristic particles per milligramme of the latter substance.

In order to determine the purity of this substance, one next prepares a mixture containing known proportions of this material and the impurities that have been found. This standard mixture is then mixed in a known proportion with lycopodium and made into a suspension as for the original powder and counts are made as explained above. From these counts one finds the number of characteristic particles per milligramme of the pure substance when counted in admixture with the impurities present and this is used as the standard figure from which to calculate the amount of pure substance present in the original article.

It will be seen that the function of the lycopodium is to enable one to know in what weight of material the characteristic particles have been counted and this makes it unnecessary to use special counting chambers and makes it possible to carry out the work upon any microscope with any convenient combination of lenses.

Further details and examples will be found in the author's publications[18, 19] referred to in the bibliography.

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THE CHEMISTRY OF THE VOLATILE OIL OF MILFOIL.*

A Study of the Application of Modern Organic Chemistry to Drug Plant Investigations.† **

BY ROLAND E. KREMERS.

In recent years the experimental culture of drug plants on a semi-commercial scale has reëmphasized the fact that ultimate success depends upon the rational application of the modern sciences, and in particular has opened an attractive field for applied chemistry. Plant chemistry, or phytochemistry, as it is sometimes called, is a composite study, requiring in its entirety a knowledge not merely of chemistry, but of botany, pharmacy, pharmacology, physiography, climatology, soils, and many other phases of science. Since no individual can devote special attention to all of these relations it is the more necessary for those who study the various phases of the subject to explain to their associates their methods and results. Hence it has seemed more rational to present the recent researches on the oil of milfoil from the point of view of the organic chemist, because that has been the training of the writer.

Although milfoil, botanically—Achillea millefolium—was known to Dioscorides[1] and has enjoyed a varying popularity as a remedy through all ages, and in spite of the fact that the first chemical examination was made by Bley[2] as far back as 1828, Miller[3] wrote in 1916 that up to that time only two constituents had been definitely identified—acetic acid and cineol. He himself, however, by a very painstaking investigation, carried out while chemist of the Wisconsin Pharmaceutical Experiment Station, was able to add very materially to the list of known constituents of this oil; and the work of the present investigator has been in continuation of his. As it is not so much the object of this paper to de-

* Contribution from the Organic Laboratory, Vanderbilt University, in coöperation with the Wisconsin Pharmaceutical Experiment Station.

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[1] Dioscorides, Materia Medica, cf. Miller, p. 5.

[2] L. F. Bley, *Trommsdorf's Neues Jour. d. Pharm.*, 16, I, pp. 245–73 (1828); and *Ibid.*, 16, II, pp. 94–120 (1828); cf. Miller, p. 7.

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