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HOW OIL AND VITAMIN A ARE DETERMINED IN FISH LIVERS

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With an increased volume of livers being sold on a potency basis, those fishermen, dealers, and others not familiar with laboratory techniques are becoming interested in the methods used by the chemists to determine oil content and vitamin A potency.

The procedures, particularly the determination of oil content, while requiring technical skill and training, are relatively simple. A sample of liver is weighed, a measured volume of a solvent similar in nature to gasoline is added, and the mixture is shaken. During this shaking, the oil and vitamin A in the liver are dissolved in the solvent. Then the undissolved liver material and the water are allowed to settle; leaving a clear solution of oil and vitamin A in the solvent. (Usually a centrifuge is used to hasten this separation.) Next a measured portion of this clear solution is placed in a container and heated slightly to drive off the solvent; after which the oil which remains is weighed. Knowing (1) the weight of oil dissolved in a measured portion of the solution, (2) the total volume of solvent used to extract the oil, and (3) the weight of liver sample from which the oil was extracted, the percentage by weight of oil in the liver can be calculated.

A second measured portion of the clear solution is used for the determination of vitamin A 1/. This measurement depends essentially upon the fact that vitamin A absorbs ultraviolet light and the more vitamin A there is in the solution, the more light that solution will absorb. That is, if an attempt is made to transmit ultraviolet light through a solution of vitamin A, the more vitamin A present, the less light will pass through.

^{1/} At the present time, the only official method for the determination of vitamin A is a biological test according to a procedure promulgated by the United States Pharmacopoeia. Since this method is relatively time-consuming and expensive, most, if not all, livers bought and sold on potency are traded on the basis of a spectrophotometric test. For this reason, the biological test, although official, is not described here.

While ultraviolet light is not visible to the human eye, there are a number of other light sensitive devices such as photographic film and photoelectric cells which do not share this limitation. Perhaps the most useful of these are the photoelectric cells, as they have the very convenient property of producing an electric current which is proportional to the intensity of the light falling upon them. This current can be measured by means of an electric meter Thus, the intensity of a beam of light invisible to the eye is translated into a reading on a meter.

If a beam of white light is allowed to pass through a slit and then through a glass prism, an image of the slit can be projected onto a screen. This image will not be white, however, because the light has passed through the prism. One side of the image will be red, while the other side will be violet, with the colors orange, yellow, green, blue, and indigo between them. Now if a narrow slit is cut in the screen and a second screen placed behind the first, any color desired can be isolated on the second screen either by moving the slit in the first screen or by rotating the prism so that only the desired color is transmitted to the second screen. If a photoelectric cell is substituted for the second screen, it will be found to react even when the prism is turned beyond the violet light and nothing visible to the eye can be seen falling on the photoelectric cell. This reaction is due to the ultraviolet light.

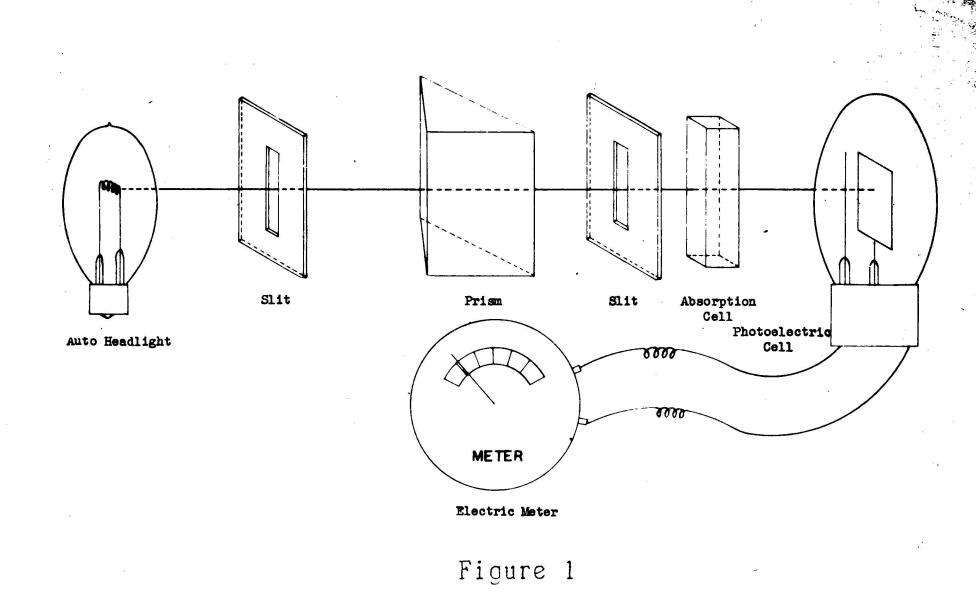
These principles are utilized in the measurement of vitamin A by means of the spectrophotometer. A schematic drawing of the operating principles of this instrument is presented in Figure 1. An ordinary automobile headlight serves as the source of illumination. A slit placed between the headlight and the prism isolates a narrow beam of light, which is separated into the various colors of the spectrum when passed through the prism. The prism is rotated in such a way that the particular color desired (ultraviolet light for vitamin A studies) falls upon a second slit. This light then passes through a glass cell containing the solution of vita min A, and, in turn, falls upon the sensitive element of a photoelectric cell. An electric meter connected to the photoelectric cell then indicates the amount of ligh passed by the vitamin A solution.

By first using oils of <u>known</u> potencies, the readings of the electric meter corresponding to various amounts of vitamin A can be determined. Thereafter, when an oil solution of <u>unknown</u> vitamin A potency is placed in the glass cell, its value can readily be determined from the reading of the electric meter. 2/

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^{2/} The values obtained by this method are called spectrophotometric or "spec" units These are not to be confused with U.S.P. units which are based upon a determination by means of the biological test. (See footnote 1)



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