given and the weight increase obtained, whether the carotene be given in one dose or in divided doses. 0.005 mg. allows a growth of at least 2 gm., the average growth increment being 3 to 5 gm. for this amount. This corresponds closely to Sherman's vitamin A unit.

The rate of fading of carotene dissolved in different solvents was determined by comparing 0.017% solutions with a standard $K_2Cr_2O_7$ solution colorimetrically. Carotene fades very rapidly in ethyl oleate, less in ethyl laurate. The color is fairly stable in ethyl oleate with hydroquinone (0.1%), and disappears most slowly in cottonseed oil. If a synthetic solvent is to be used, ethyl laurate protected with hydroquinone seems the logical choice. Whether the fading in solution is comparable to the fading of the crystals, is uncertain, but in either case, the physiological activity is lost.

Carotene is a pro-oxidant. 0.06% dissolved in an autoxidizable oil shortens its induction period by one-half. It is possible that this property is closely allied to its function as vitamin A.

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Determination of Butyl and Ethyl Alcohols in Fermentation Mixtures.

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An accurate and rapid method for the quantitative determination of alcohols in fermentation mixtures would prove of value to the zymologist engaged in studies of the chemism of microbial action. A method which has been found very satisfactory is based upon the oxidation of the alcohols in the neutral volatile distillate by potassium dichromate and phosphoric acid. Ethyl alcohol yields acetic acid and butyl alcohol yields a mixture of butyric and acetic. The acids are quantitatively determined by the partition method.[1] The method is presented in its present form for the quantitative determination of unknown mixtures of butyl and ethyl alcohols, such as are found in the "butyl-acetone" fermentation. This fermentation has extensive industrial use in the production of butyl alcohol and acetone.

Fifty cc. of the alcohol mixture are pipetted into a 200 cc. balloon flask containing 10 gm. of potassium dichromate and 25 cc. of c.p. 85% phosphoric acid. Three or 4 small pieces of porcelain are

[1] Werkman, C. H., *Ind. Eng. Chem.* (analytical edition), 1930, **2**, 302.

added to insure smooth boiling and the flask is connected to an efficient reflux condenser. The flask is then heated at such a rate that the mixture is brought to a boil in one and one-half minutes. Gentle boiling is maintained for 3 minutes, making the total time of heating between 4 and 5 minutes. It is advisable to wash down the condenser tube occasionally by adding 5 cc. of water at the top. The volume of liquid in the flask should never exceed 100 cc.

After oxidation is complete the flask is quickly connected to a Liebig condenser and distilled until the mixture in the flask begins to foam. The heat is then lowered until foaming just continues. The heating is then continued for 2 or 3 minutes. If this procedure is carried out carefully all of the volatile acid is carried over but none of the phosphoric. The mixture has no tendency to bump or spatter.

The distillate is made up to 100 cc. and the total acid determined by titrating 25 cc. The result should be expressed in cc. of 0.1 N acid.

Thirty cc. are then partitioned with iso-propyl ether as described by Werkman.[1]

Under these conditions ethyl alcohol is quantitatively converted to acetic acid and butyl alcohol to a mixture containing 90% butyric acid and 10% acetic acid. This ratio of butyric to acetic acid was determined by oxidizing 5 butyl alcohol solutions varying in concentration from 50 mg. of alcohol to 600 mg. per 50 cc. The percentages of butyl alcohol found were 90, 90.2, 90, 90.5, and 91.

The calculations are most easily made in terms of cc. of 0.1 normal solutions.

From Werkman[1] may be read the per cent of butyric acid. This per cent divided by 0.9 and multiplied by the cc. of 0.1 N acid in the distillate gives the total cc. of 0.1 N acid derived from the butyl alcohol and designates the cc. of 0.1 N butyl alcohol in the alcohol mixture.

The difference between the total acid and that representing butyl alcohol gives the cc. of 0.1 N acid representing the ethyl alcohol. An example will make the meaning clear.

Assume that the distillate contains exactly 100 cc. of 0.1 N acid.

The partition constant is 16.5. Reference to the nomogram[1] shows that the mixture contains 60% butyric and 40% acetic acid. Then $0.60 \times 100/0.9 = 66.6$ cc. of 0.1 N acid = 66.6 cc. of 0.1 N butyl alcohol. $100 - 66.6 = 33.4$ cc. of 0.1 N ethyl alcohol.

The method was applied to 15 known alcohol mixtures. The maximum error was 3.5% for ethyl alcohol and 3.0% for butyl alcohol.