proved to be so variable, depending upon the subjects' activity, that only the mean readings of the morning (basal) pulse are shown in Table I.

There was very slight increase in the pulse rate in 7, no change in 3 and a slight decrease in 2 cases. In no individual case was any marked effect noted, neither do the means for each sex show significant change in value. The general tendency of the means for all subjects is toward an increase, but the changes are inconsequential, when tested by the criterion that the difference in two values to bear significance must be greater than 3 times the probable error.[1] From the results of this small series it is concluded that iodine in the form of Lugol's solution, 10 drops once daily, has no appreciable effect on the pulse rate of normal individuals.

[1] Pearl, Raymond, introduction to *Medical Biometry and Statistics*, 1923, Philadelphia (p. 214).

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One Solution Technique for Direct Microscopic Counting of Bacteria in Milk.

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A method has been devised for simplifying the direct microscopic method of Breed for counting bacteria in milk.

In the official Breed method, 0.01 ml. of milk is spread over an area of 1 square centimeter on a glass slide. When dry, the slide is immersed in xylene for at least one minute (to remove the fat from the milk); then dried, immersed in 90 per cent alcohol for one or more minutes (to fix the smear to the slide); dipped in Loeffler's methylene blue until the milk smear is overstained; washed in water; decolorized in alcohol; again washed in water; dried and examined under a standardized microscope.

The new method eliminates the use of two solutions, two Coplin jars and four separate operations, and substitutes a single solution which simultaneously dissolves out the fat; fixes the milk smear to the slide and stains the bacteria and leucocytes. Excellent contrast between bacteria and background is secured without recourse to decolorization. Any one of the following solutions may be used:

Formula I.

Methylene blue, pwd.,	2.0 gm.
Ethyl alcohol, 95 per cent	60.0 ml.
Xylene, cp.,	40.0 ml.
Acetic acid, glacial	6.0 ml.

Dissolve methylene blue* in warm (70° C.) alcohol, adding the powdered stain very slowly. Then add the xylene and acetic acid. Filter. Keep in tightly stoppered bottle.

1. Milk smears are made on a glass slide in the usual manner. When dry, the slide is immersed in the above solution for ½ to 4 minutes.

2. Remove the slide from the solution and allow it to drain until the *smear* is thoroughly *dry*. In warm weather, this seldom requires longer than half a minute.

3. Wash *thoroughly* in water.

Formula II.

Methylene blue*, pwd.,	1 gm.
Ethyl alcohol, 95 per cent	54 ml.
Tetrachlorethane, tech.†	40 ml.
Acetic acid, glacial	6 ml.

Add the alcohol to the tetrachlorethane in a flask and bring to a temperature not to exceed 70° C.; add to the powdered methylene blue. Shake vigorously until the dye is completely dissolved; then add the glacial acetic acid. Filter the entire volume through a 15 cm. filter paper. Keep in tightly stoppered bottle.

Directions or Use.

1—Prepare milk smear. When dry
2—Immerse the smear in the solution and withdraw immediately.
3—Drain until dry (about 30 seconds).
4—Wash in water.
5—Dry and observe.

Formula III.

Methylene blue,* pwd.	0.7 gm.
Ethyl alcohol, 95 per cent	30.0 ml.
Tetrachlorethane, tech.†	40.0 ml.
Pyridine	15.0 ml.
Amyl acetate	15.0 ml.

* Certified.

† Can be obtained from Eastman Kodak Co. Technical tetrachlorethane is relatively inexpensive.

Add the warm mixture of alcohol and tetrachlorethane to the powdered methylene blue and shake vigorously until dissolved. When cool, add the pyridine and amyl acetate and filter. Keep in tightly stoppered bottle.

The same staining directions are followed in using this last solution as are given under Formula II.

Formula III, which is an alkaline solution, is submitted to meet any possible objection to the use of an acid-methylene blue stain. Pyridine is incorporated as an accessory fixative agent. The function of the amyl acetate is to partially mask the odor of pyridine.

A series of comparative counts made in the laboratory on raw and pasteurized milk of high and low count revealed no apparent variation from the official Breed method.

Although the use of Formula I has been found very satisfactory, it is believed that because of the added advantages possessed by Formulae II and III, they will be found more satisfactory and for this reason are recommended.

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Presence of Physiologically Active Substance in Two California Species of Ephedra.

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Current interest in Ma Huang,[1] the Chinese drug plant *Ephedra,* makes imperative a study of the various species of the genus, especially with respect to the activity and nature of the chemical principles which they yield.

The problem is complicated by the lack of knowledge concerning the effect of species variation and ecological conditions upon the yield of physiologically active substances.

A complete study involves: (1) Determination of the species and varieties which contain physiologically active substances. (2) The isolation and identification of the active principles. (3) The experimental and clinical testing of the pure principles obtained.

In the work reported in this paper our efforts have been confined to a comparative study of the activity of *Ephedra californica*