An Enzymatic Spectrophotometric Method for the Determination of Xanthine and Hypoxanthine

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A method for determining the total content of the oxypurines xanthine and hypoxanthine in urine is described which is technically simplified and has greater specificity than existing methods. This method is highly specific, reproducible, and accurate. It also allows an estimation of the present xanthine and hypoxanthine in the urine without performing any additional measurements. When applied to plasma, special concentrating procedures are required and the method is therefore less precise.

Interest in determining the content of xanthine and hypoxanthine in biologic fluids has been stimulated by the clinical use of the drug allopurinol‡ (4-hydroxypyrazolo [3,4-d] pyrimidine), a potent inhibitor of xanthine oxidase. This compound has been used in cancer chemotherapy to enhance the action of certain drugs that are known to be degraded by xanthine oxidase (1). It has also provided a new approach to the treatment of gout through its inhibition of uric acid production. The oxypurines xanthine and hypoxanthine appear in increased quantities in the urine and serum as a consequence of administration of allopurinol (2).

Colorimetric (3), chromatographic (4), and enzymatic spectrophotometric (5) methods have been developed for the determination of oxypurines. The latter method, as described by Kalckar (5), is based on the change in absorption spectrum that accompanies the oxidation of hypoxanthine or xanthine to uric acid. Jorgensen and Poulsen (6) and Petersen et al. (7) have adapted the enzymatic procedure to use with human plasma and urine.

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The present method for determining total oxypurines* in urine is technically much easier to perform and has greater specificity than previous methods (6, 7). It avoids the necessity of taking absorbance (A) readings at two different wave lengths and also avoids radical changes in buffer pH throughout the course of the reactions. This method permits the determination of total oxypurines with the same high degree of specificity already demonstrated for uric acid by the use of uricase (8). With relatively simple additional calculations, an estimate of the percent xanthine and hypoxanthine in the urine can also be obtained. Modifications of this same method are described for determination of total oxypurine content in plasma.

**Principle**

The change in absorption spectrum accompanying the conversion by xanthine oxidase of hydroxanthine or xanthine in alkaline solutions to uric acid results in an increased absorbance at 292 nm (Fig. 1), while the enzymatic oxidation of uric acid by uricase results in a decreased absorbance at 292 nm. These principles have previously been used for the determination of uric acid (5, 8), and oxypurines (5–7). By combining these methods it is possible to determine the quantity of uric acid in a biologic fluid before (preformed uric acid) and after (total uric acid) treatment with xanthine oxidase. The difference represents the oxypurine (hypoxanthine and xanthine) content. The relative contribution of xanthine and of hypoxanthine to total oxypurines can also be determined by making use of the fact that xanthine has a substan-

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*For the purpose of clarity in this paper the term “total oxypurines” will refer only to hypoxanthine and xanthine, even though uric acid is also an oxypurine.
tially greater absorbance at 292 nm than does hypoxanthine (Fig. 1).

In order to assay for oxypurines in human plasma, some modification of the procedure is required. The very low concentration of oxypurine in plasma cannot be determined in the presence of the relatively large amounts of protein and uric acid present in plasma. This problem can be overcome by destroying the preformed uric acid by treatment with uricase and by removing the proteins. The uricase is then destroyed by addition of strong alkali (9) which is then neutralized by acid and the determination made as above. Because of the high absorbance at 292 nm contributed by plasma proteins, oxypurines are separated from proteins of plasma by dialysis and concentrated by lyophilization of the dialysate to dryness.

**Method**

**Materials**

1. *Xanthine and hypoxanthine (chromatographically pure)* Obtained from Schwarz Bio-Research, Inc., Orangeburg, N. Y.
4. *Parafilm (Grade M)* Obtained from Marathon Corp., Menasha, Wis.
5. *Purified uricase (0.75 units/ml.)* Obtained from Worthington Biochemical Company, Freehold, N. J. A unit of activity is that amount which oxidizes 1 μmole of uric acid per minute to allantoin at 25° in 0.1 M borate buffer (pH 8.5) containing 20 μg. uric acid per milliliter.
6. *Milk xanthine oxidase (9 units/ml.)* Obtained from Worthington Biochemical Company, Freehold, N. J. A unit is that quantity forming 1 μmole of uric acid per minute at 25° from 20 μg. hypoxanthine per milliliter of 0.05 M phosphate buffer at pH 7.5. An aliquot of the xanthine oxidase was diluted with 2 volumes of 0.1 M (pH 8.0) pyrophosphate buffer prior to use.

Some recent lots of commercial xanthine oxidase have a greater specific activity and therefore need further dilution. It has also been noted when using the more concentrated xanthine oxidase preparations that there is a gradual decrease in the absorbance at 292 nm in the reaction cuvets after the addition of xanthine oxidase, suggesting that the newly formed uric acid is being destroyed. These preparations are free of contaminating uricase and therefore this loss of uric acid is the result of nonenzymatic uricolyis, probably due to hydrogen peroxide accumulation. Presumably less pure xanthine oxidase prepara-
tions contain catalase which prevents this reaction. Therefore, the addition of 0.1 ml. 2% mercaptoethanol or 0.1 ml. purified catalase to each 1 ml. of diluted xanthine oxidase is suggested if there is any evidence of nonenzymatic breakdown of uric acid in the reaction cuvet.

Apparatus

All determinations were made with a Beckman DU spectrophotometer equipped with an ultraviolet attachment, and 4.0-ml. capacity silica cuvets with a 1.0-cm. light path. Pipets of a 0.1-ml. capacity and a graduation interval of 0.001 ml. for pipetting the enzymes were also required.

Serum dialysates were lyophilized using a Virtis freeze dryer (Virtis Company, Inc., Yonkers, N.Y.).

Reagents

Pyrophosphate buffer (0.1 M, pH 8.0) and glycine buffer (0.1 M, pH 9.4) were selected for the pH optimum of xanthine oxidase (10) and of uricase (5), respectively. However, the rate of reaction of uricase in pyrophosphate buffer at pH 8.0 is sufficiently great to allow the reaction to go to completion when an excess of uricase is used. Since uric acid has the same molar absorbance index at pH 8.0 and pH 9.4 the same methods of calculation can be performed regardless of which buffer is used in the reaction mixture.

Procedure

Total Oxypurine Concentration in Urine

Total oxypurines = total uric acid — preformed uric acid.

Urine is collected with 3 ml. toluene as a preservative and stored at room temperature to prevent the precipitation of any uric acid or oxypurine. The usual period of urine collection is 24 hr.

After appropriate dilution with water (usually in the range of 1:50 to 1:200) the sample can be stored in a tightly sealed container at 5° for at least 21 days without significant change in content of uric acid or oxypurines.

Preformed Uric Acid

This is determined by the method of Liddle et al. (7). Duplicate assay cuvets each contain 2.0 ml. of 0.1 M glycine pH 9.4 and 1.0 ml. of urine diluted sufficiently to give an absorbance at 292 nm of approximately 0.200. A cuvet containing 3.0 ml. of the glycine buffer serves as an enzyme blank. Under exceptional circumstances a third cuvet—a biologic fluid blank containing only urine and buffer but no enzyme
will be required to correct for spontaneous changes in absorbance that are induced by the alkali. The absence of such a blank change in normal urine permits its omission from routine determinations. The reference cuvet contains water. After determining the initial absorbance at 292 nm, 0.01 ml. of undiluted purified uricase is added to each cuvet and the solutions are mixed by covering the cuvets with parafilm and inverting them several times. The reaction is allowed to proceed until there is no further change in the absorbance in successive 5-min. readings (usually about 30 min.). This reading becomes the final absorbance.

**Calculations**

\[
\frac{\text{\textmu} \text{mole uric acid}}{\text{Ml. urine}} = \frac{\Delta A \text{ preformed uric acid}}{12.5 \text{ A/\textmu mole/ml.}} \times 3.01 \times \text{dilution of urine}
\]

\(\Delta A\) preformed uric acid is the difference between the initial absorbance and the absorbance after reaction with uricase (after correcting for the small changes observed in the enzyme blank). The 12.5 A/\textmu mole/ml. is the molar absorbance index for uric acid.

**Total Uric Acid**

This determination is performed in two steps with readings at 292 nm as follows:

<table>
<thead>
<tr>
<th>Oxypurines</th>
<th>xanthine</th>
<th>uric acid</th>
<th>uricase</th>
<th>allantoin</th>
</tr>
</thead>
<tbody>
<tr>
<td>(Initial A)</td>
<td>oxidase</td>
<td>(intermediate A)</td>
<td>(final A)</td>
<td></td>
</tr>
</tbody>
</table>

Reading I | Reading II | Reading III

Preferably, the same dilution of urine as for the previous determination will be used. However, if the total oxypurine content is small (less than 0.06 mM/24 hr.), a more concentrated solution may be necessary.

Duplicate assay cuvets each containing 1.0 ml. of diluted urine and 2.0 ml. of 0.1 M pyrophosphate buffer pH 8.0 are prepared. An enzyme blank and a reference cuvet containing water are used. The initial A at 292 nm is determined (Reading I) and then 0.01 ml. of the 1:3 diluted milk xanthine oxidase is added to each cuvet. The cuvets are mixed and the reaction is allowed to proceed until there is no further change in the absorbance (usually about 10 min.). If the xanthine oxidase inhibitor allopurinol has been administered to the patient whose urine is being analyzed, larger quantities of xanthine oxidase (up to 0.04 ml.) may be required to achieve complete reaction in a reasonable period of time, or the urine may need to be diluted further to diminish concentrations of inhibitor. After the final absorbance for this reaction is determined (Reading II), 0.01 ml. of purified uricase
is added to each cuvet, the cuvets are mixed, and the reaction allowed to go to completion (about 30 min.). The final absorbance for the uricase reaction is then determined (Reading III). The ΔA for total uric acid is then calculated (Reading II minus Reading III) making appropriate corrections for changes in the absorbance of the enzyme blank and reference cuvet.

The presence of inhibitors of xanthine oxidase (such as allopurinol and its metabolites) in the fluids being assayed could result in incomplete conversion of oxypurines to uric acid. The complete recovery in the assay system of a known quantity of xanthine added to a duplicate sample of the fluid being assayed provides proof that an excess of xanthine oxidase has converted all oxypurines to uric acid.

**Total Oxypurines**

ΔA of uric acid derived from oxypurine = ΔA total uric acid minus ΔA preformed uric acid.

**Calculations**

\[
\frac{\mu M \text{ total oxypurine}}{\text{Ml. of urine}} = \frac{\Delta A \text{ uric acid derived from oxypurine}}{12.5 \text{ A/}\mu M/\text{ml.}} \times 3.01 \text{ ml.} \times \text{dilution of urine}
\]

**Percent Xanthine and Hypoxanthine**

The increase in absorbance at 292 nm accompanying the oxidation of 1.0 μM of xanthine per milliliter to uric acid is 9.29, while that for oxidation of hypoxanthine is 12.33, a difference of 3.04 A/μM/ml.

ΔA disparity due to xanthine = ΔA of uric acid derived from oxypurine (see Total Oxypurines above) — ΔA of assay cuvet after xanthine oxidase alone (Reading II — Reading I).

**Calculations**

\[
\frac{\mu M \text{ xanthine}}{\text{ML of urine}} = \frac{\Delta A \text{ disparity}}{3.04 \text{ A/}\mu M/\text{ml.}} \times \frac{3.01 \text{ ml.}}{1 \text{ ml.}} \times \text{dilution of urine}
\]

Percent of total oxypurine which is xanthine or hypoxanthine:

\[
% \text{ xanthine} = \frac{\mu M \text{ xanthine}/\text{ml.}}{\mu M \text{ total oxypurine}/\text{ml.}}
\]

\[
% \text{ hypoxanthine} = 100 - % \text{ xanthine}
\]

**Example of Calculations**

24-hr. urine volume = 2000 ml.
Urine diluted 1:100 with water.

**Preformed Uric Acid**

Add 1 ml. of diluted urine and 2 ml. of 0.1 M glycine buffer pH 9.4 to duplicate cuvets.
Initial absorbance at 292 nm is 0.235.
Add 0.01 ml uricase and allow reaction to go to completion (i.e.,
until no further changes in absorbance occurs over a 5-min. interval).
Final absorbance = 0.135.

\[ \Delta A \text{ preformed uric acid} = 0.235 - 0.135 = 0.100 \]

\[ \frac{\mu M \text{ preformed uric acid}}{\text{Ml. urine}} = \frac{100}{12.5} \times 3.01 \times 100 = 2.408 \]

Molecular weight of uric acid = 168.

\[ \frac{\text{Mg. preformed uric acid}}{24 \text{ hr.}} = \frac{\mu M \text{ uric acid}}{\text{ml. urine}} \times \frac{\text{mM}}{1000 \mu M} \times \frac{168 \text{ mg.}}{\text{mM}} \times \frac{\text{vol. (ml.)}}{24 \text{ hr.}} \]

\[ \frac{\text{Mg. preformed uric acid}}{24 \text{ hr.}} = 2.408 \times \frac{1}{1000} \times 168 \times 2400 = 809 \]

**Total Uric Acid**

Add 1 ml of the same diluted urine and 2 ml of 0.1 M pyrophosphate
buffer pH 8.0 to duplicate cuvets.
The initial absorbance at 292 nm is recorded, e.g., 0.235. Then 0.01 ml
xanthine oxidase is added to each cuvet and the reaction is allowed to
go to completion.
The absorbance is again recorded (the intermediate A) e.g., 0.300.
Then 0.01 ml uricase is added to each cuvet and the final A recorded,
e.g., 0.125:

\[ \Delta A \text{ total uric acid} = 0.300 - 0.125 = 0.175 \]

**Total Oxypurines**

\[ \Delta A \text{ uric acid derived from oxypurines} = \Delta A \text{ total uric acid minus} \]
\[ \Delta A \text{ preformed uric acid.} \]

\[ \Delta A \text{ uric acid derived from oxypurines} = 0.175 - 0.100 = 0.075 \]

\[ \frac{\mu M \text{ total oxypurines}}{\text{Ml. urine}} = \frac{0.075}{12.5} \times 3.01 \times 100 = 1.806 \]

\[ \frac{\text{Mg. uric acid derived from oxypurines}}{24 \text{ hr.}} = 1.806 \times \frac{1}{1000} \times 168 \times 2400 = 727 \text{ mg.} \]

**Percent Xanthine and Hypoxanthine**

The \( \Delta A \) due to the addition of xanthine oxidase is the difference
between the intermediate A and the initial A.

\[ \Delta A = 0.300 - 0.235 = 0.065 \]

The \( \Delta A \) uric acid derived from oxypurines was previously calculated
as the difference between $\Delta A$ total uric acid and $\Delta A$ preformed uric acid.

$\Delta A$ uric acid derived from oxypurines, (0.075) and $\Delta A$ due to the addition of xanthine oxidase (0.065):

$$\Delta A \text{ discrepancy due to xanthine} = 0.075 - 0.065 = 0.010$$

$$\frac{\mu M \text{ xanthine}}{\text{ml urine}} = \frac{0.010}{3.04} \times 3.01 \times 100 = 0.99$$

Percent xanthine = $$\frac{\mu M \text{ xanthine/ml urine}}{\mu M \text{ total oxypurine/ml urine}} = \frac{0.99}{1.806} = .553$$

Therefore, there is 55.3% xanthine and 44.7% hypoxanthine in this example.

**Total Oxypurine Concentration in Plasma**

Blood is drawn into a heparinized syringe, transferred to a test tube, and immediately chilled in ice to prevent the erythrocytes from forming oxypurines (11). Plasma is removed by centrifugation in a refrigerated centrifuge. No change in oxypurine content of heparinized blood was found in 30 min. at 0$^\circ$.

Plasma (4–12 ml.) is placed in collodion dialysis tubing and is dialyzed at 24$^\circ$ against 5 volumes (20–60 ml.) of distilled water with constant agitation for 6 hr. The volume of the dialysate is measured, lyophilized, and the dry powder resuspended in 6.0 ml. of 0.1 M pyrophosphate buffer pH 8.0 in a 10-ml volumetric flask. To destroy all uric acid present, 0.03 ml. of purified uricase is added and is allowed to react for about 12 hr. at 24$^\circ$. The uricase is then destroyed by the addition of 0.25 ml. of 5 N NaOH. After 20 min. the pH is adjusted back to 8.0 with 5 N HCl. Then 0.1 M pyrophosphate buffer pH 8.0 is added to bring the total volume to 10 ml.

A 3-ml aliquot of the solution is placed in triplicate assay cuvets. In another cuvet, 3 ml. of 0.1 M pyrophosphate buffer pH 8.0 is used as an enzyme blank. Mild xanthine oxidase (0.01 ml.) is added to the 4 cuvets and the solutions are mixed. The reaction is allowed to proceed to completion, converting all of the oxypurine to uric acid. The uric acid is then destroyed by addition of 0.01 ml. purified uricase to two assay cuvets and then to the enzyme blank. Again the reaction is permitted to go to completion (about 30 min.) and the final absorbance is read at 292 nm. No uricase is added to the third assay cuvet which serves as the biologic fluid blank. Absorbance readings for this cuvet are taken at the same times as the assay cuvets and the enzyme blank.
Calculations

\[
\frac{\mu M \text{ of total oxypurine}}{\text{Ml. of plasma}} = \frac{\Delta A \text{ uric acid derived from oxypurine}}{12.5 \text{ A}/\mu M/\text{ml.}} \times \frac{3.03}{3} \times 10 \times \frac{6 \text{-fold dilution}}{\text{vol. of dialysate lyophilized}}
\]

\[
\frac{\mu M \text{ total oxypurine}}{\text{Ml. of plasma}} = \frac{\Delta A \text{ uric acid} \times 4.85}{\text{vol. of dialysate lyophilized}}
\]

Results and Discussion

Specificity

Although xanthine oxidase reacts with many different purines and aldehydes, uricase is highly specific for uric acid. Since hypoxanthine and xanthine are the only naturally occurring purines that form uric acid upon oxidation by xanthine oxidase, the determination of total oxypurines as outlined here has a correspondingly high degree of specificity. The oxidation of 1-methyl xanthine catalyzed by xanthine oxidase results in the formation of 1-methyl uric acid which has an absorption spectrum similar to that of uric acid (12). However, the 1-methyl uric acid fails to react with uricase and therefore does not interfere in the determination of total oxypurines. It does interfere with the procedure for determining the percent xanthine or hypoxanthine. Thus, ingestion by a patient of beverages containing methyl-
activated purines may result in an apparent xanthine content in excess of 100% (Fig. 2) and therefore precludes the determination of percent xanthine in the urine. However, since methylated purines are not affected by uricase, they do not interfere with the determination of the total oxypurine content of urine or plasma by this method. Likewise, allopurinol does not interfere with the determination of the total oxypurine content of urine or plasma as long as the precautions are taken as described to assure an excess of xanthine oxidase activity in the reaction mixtures. Theoretically, allopurinol administration could alter the determination of the percent xanthine since the oxidation product of allopurinol, dihydrooxypyrarzolo pyrimidine, has a greater absorbance at 292 nm than the parent compound. In practice, however, the quantities of those compounds present in biologic fluids and the relative slowness of this oxidation reaction makes this of little concern and does not justify the added difficulties and inaccuracies introduced by chromatographic separation of these compounds.

Reproducibility

The reproducibility for uric acid determination by the uricase method has previously been determined (8).

The reproducibility for total oxypurine determination in the urine and plasma is shown graphically in Fig. 3 and 4. The standard deviations* of duplicates for each group of analyses were ± 0.01 mM/24 hr. for urine, determined from 50 paired observations (range 0.01–4.71 mM/24 hr.) and ± 0.02 μmole/100 ml. for plasma, determined from paired observations (range 0.09–4.06 μmole/100 ml.).

The reproducibility for the percent xanthine in the urine is shown in Fig. 2. The standard deviation in 31 duplicate determinations (range 0–160%) was ± 20%.

Accuracy

The accuracy of the method for determining total oxypurine and percent xanthine and hypoxanthine in buffered solutions also containing uric acid is demonstrated by the recovery of oxypurines shown in Table 1. The accuracy was greatest for determination of total oxypurines in urine, which is the primary purpose for which this method was devised. Determination of the percent xanthine in urine and of the total oxypurine determination in plasma were subject to much greater variability but may still be of value for many types of clinical

\[ \text{Standard deviation} = \pm \sqrt{\frac{\text{sum of } D^2}{2N}} \]  
where $D =$ difference between duplicate determination and $N =$ number of pairs of determinations (15).
investigations. The data in Table 1 indicate that the determination of the percent xanthine in known mixtures averaged 7–10% higher than was actually present.

The presence of a xanthine oxidase inhibitor in the urine or plasma being assayed may lead to some difficulties in performing these determinations. Since one of the chief uses of this method is to monitor the clinical effectiveness of the xanthine oxidase inhibitor allopurinol, special care must be taken to overcome this difficulty. After the addition of xanthine oxidase, it is necessary to be sure the reaction is complete before taking final readings or adding the final uricase. The most

Fig. 3. Reproducibility of enzymatic spectrophotometric method for determination of total oxypurines in urine. Points represent duplicate analyses.

Fig. 4. Reproducibility of method for determination of total oxypurines in plasma.
<table>
<thead>
<tr>
<th>Uric acid (mg./100 ml.)</th>
<th>BUFFER SOLUTIONS CONTAINING ADDED URIC ACID</th>
<th>Xanthine (% of total oxypurine)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Added</td>
<td>Found</td>
</tr>
<tr>
<td>7.6</td>
<td>86.2</td>
<td>86.5</td>
</tr>
<tr>
<td>7.6</td>
<td>90.5</td>
<td>93.8</td>
</tr>
<tr>
<td>3.0</td>
<td>48.1</td>
<td>50.1</td>
</tr>
<tr>
<td>4.3</td>
<td>3.3</td>
<td>76.7</td>
</tr>
<tr>
<td>12.1</td>
<td>15.9</td>
<td>15.9</td>
</tr>
<tr>
<td>0.3</td>
<td>70.5</td>
<td>70.0</td>
</tr>
<tr>
<td>0</td>
<td>31.8</td>
<td>31.3</td>
</tr>
<tr>
<td>0</td>
<td>58.4</td>
<td>58.6</td>
</tr>
<tr>
<td><strong>AVERAGE</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>RANGE</strong></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

| URINE | | | | | |
|-------|----------------|----------------|
|       | Add     | Found  | % Found | Add     | Found  | % Found |
| 8.4   | 87.1    | 88.4   | 102.6   | 26.1    | 36.8   | 141.0   |
| 16.7  | 91.5    | 94.4   | 103.2   | 41.2    | 47.8   | 116.0   |
| 16.7  | 16.0    | 15.1   | 94.4    | —       | —      | —       |
| 8.4   | 49.0    | 49.4   | 100.8   | 77.0    | 85.3   | 110.8   |
| 16.7  | 33.9    | 30.0   | 88.5    | 1.5     | 1.0    | 66.7    |
| 0.4   | 3.0     | 3.0    | 100.0   | 98.8    | 102.0  | 103.2   |
| **AVERAGE** |       |       |         | 98.2    |       | 107.5   |
| **RANGE** |       |       |         | 88.5-103.2 |       | 66.7-141.0 |

| PLASMA | | | | | |
|--------|----------------|----------------|
|        | Add     | Found  | % Found | Add     | Found  | % Found |
| 3.48*  | 3.13    | 89.9   |         |         |        |        |
| 6.68†  | 6.20    | 92.8   |         |         |        |        |
| 5.26*  | 5.15    | 97.9   |         |         |        |        |
| 11.82* | 11.25   | 95.2   |         |         |        |        |
| 4.86†  | 4.80    | 98.8   |         |         |        |        |
| 14.66† | 14.16   | 96.6   |         |         |        |        |
| **AVERAGE** |       |       |         | 95.2    |       |        |
| **RANGE** |       |       |         | 89.9-98.8 |       |        |

* Xanthine added.
† Hypoxanthine added.

A satisfactory way to assure an excess of xanthine oxidase is to add a known amount of xanthine to the cuvet at completion of the reaction, allow the added xanthine to be oxidized, and show that the appropriate increase in absorbance at 292 nm occurs. If an excess of xanthine oxidase is not present, more may be added until an excess is obtained. It is not a sufficient check to add more xanthine oxidase until there is no further change in absorbance since a large excess of inhibitor may prevent the additional xanthine oxidase from reacting.

The biologic fluid blank corrects for any spontaneous changes in absorbance of plasma during the reaction period. This is not necessary...
in urine determinations except under special circumstances where unusual drugs are being administered. The need for this can be ascertained by noting any change in absorbance in the assay cuvet over a period of time (about 1 hr.) before adding the appropriate enzymes.

Normal Urinary Oxypurine Excretion

The mean 24-hr. urinary total oxypurine excretion for six normal subjects was 75 \( \mu M \) (range 30–140 \( \mu M \)). This is equivalent to approximately 11 mg. of hypoxanthine (range 4–19 mg.) or 12 mg. of xanthine (range 5–21). No attempt was made to determine the percent xanthine in these subjects, because of the low oxypurine content normally present in urine.

Normal Plasma Oxypurine Concentration

The mean plasma oxypurine concentration in four normal subjects was 0.80 \( \mu M/100 \text{ ml.} \) (range 0.48–1.25 \( \mu M/100 \text{ ml.} \)). This compares to a range of normal plasma oxypurine values from 0.7 to 2.0 \( \mu M/100 \text{ ml.} \) (0.1–0.3 mg./100 ml.) as determined by Jorgensen and Poulsen (6).

References