
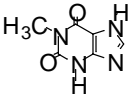
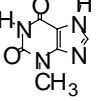
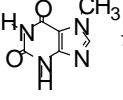
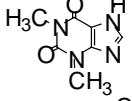
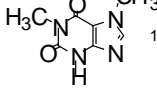
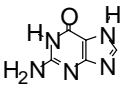
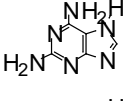
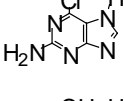
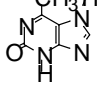
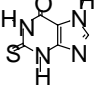
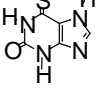


Table S2. Kinetic parameters for the reductive half-reaction of xanthine oxidase with purine substrates. For faster substrates, the anaerobic reaction of enzyme with substrate in pseudo-first order excess was followed at 450 nm (following the bleaching of enzyme upon reduction) by stopped-flow spectrophotometry, with the data were fit to single exponentials to obtain k_{obs} . For slower substrates, the reaction was followed in a diode array spectrophotometer, with rate constants obtained from linear fits to a semilogarithmic plot of $\ln(\Delta A/\Delta A_0)$ versus time. The reaction conditions were 0.1 M MES buffer, 0.1 N KCl pH 7.0, 25 °C. Plots of k_{obs} versus [substrate] were fitted using the hyperbolic relationship $k_{obs} = k_{red}[S]/(K_d + [S])$ to obtain the limiting rate of reduction, k_{red} , and substrate dissociation constant, K_d [4c]. (b) In the case of 2-hydroxy-6-methylurine, 2-thioxanthine and 6-thioxanthine there was no substrate concentration dependence observed in the reductive half-reaction. For these three substrate the relevant kinetic parameters given in Table 2 were followed under conditions of a pseudo first-order excess of enzyme rather than substrate under aerobic conditions, following the reaction by the small spectral changes arising from the molybdenum center itself.

| | | $k_{red} (s^{-1})$ | $K_d (\mu M)$ | $k_{red}/K_d (M^{-1}s^{-1})$ |
|---|--|--------------------|----------------------|------------------------------|
|  | xanthine | 7.0 | 0.54 | 1.3×10^7 |
|  | 1-methylxanthine | 13.9 | 4.5 | 3.1×10^6 |
|  | 3-methylxanthine | (0.0005) | no reaction detected | |
|  | 7-methylxanthine | — | — | 2.0×10^3 |
|  | 1,3-dimethylxanthine (theophylline) | (0.00005) | no reaction detected | |
|  | 1,7-dimethylxanthine | 0.070 | 1200 | 56 |
|  | guanine | 0.0001 | 59 | 1.7 |
|  | 2,6-diaminopurine | 0.001 | 52 | 17 |
|  | 2-amino-6-chloropurine | 0.013 | 133 | 100 |
|  | 2-hydroxy-6-methylxanthine | 0.133 | 1.15 | 1.2×10^5 |
|  | 2-thioxanthine | 3.0 | — | — |
|  | 6-thioxanthine | 1.4 | 0.95 | 1.5×10^6 |