filled by all ligands results, respectively, in m equations of type (2) and n equations of type (3):

$$C_j = N_j \sum_{i=1}^{n} K_{ij} (S_i - D_i) \Big/ \left[1 + \sum_{i=1}^{n} K_{ij} (S_i - D_i) \right] \quad (2$$

$$D_i = S_i \sum_{j=1}^{m} K_{ij} (N_j - C_j) \Big/ \left[1 + \sum_{j=1}^{m} K_{ij} (N_j - C_j) \right] \quad (3)$$

which constitute a system of $m + n$ nonlinear equations in $m + n$ unknowns. No quantity in equations (2) and (3) can be negative and, since the total concentration of a species exceeds the concentrations of its bound fraction, $N_j > C_j$ and $S_i > D_i$. These consideration and the symmetry of equations (2) and (3) permit the application of the following iterative method by successive approximations: Let X_j be an initial guess or approximation of C_j. Substitution of the m approximations of this type in the n equations (3) yields the n approximations, Y_i, for D_i. Their substitution in the m equations (2) then yields a second set of m approximations for X_j. This process is repeated until the following condition is satisfied:

$$\left| \sum_{i=1}^{n} Y_i - \sum_{j=1}^{m} X_j \right| < \delta \quad (4)$$

where δ, the tolerance, is positive and smaller than the least significant digit of accuracy desired. Substitution of the solutions in (1) gives all individual values for B_{ij}.

A program based on this algorithm was written for use on the Hewlett-Packard model 9810A calculator. The program consists of 1123 steps. The storage capability of this calculator (111 data registers in option 001) places ceilings for m and n which must satisfy the expression $mn + 2(m + n) \leq 95$. A listing of the program and instructions for use will be available from the author on request. The printed solution consists of the concentration of filled binding sites and percent occupancy for each class of sites; the concentration of bound ligand, percent total bound, and the ratio bound/unbound for each ligand; and all the individual values of B_{ij}. The method of iteration is not greatly affected by the values of the initial guesses which in the program are $N_j/2$. Using the association constants and concentrations for the binding of 6 steroid hormones and 3 plasma proteins presented by FELDMAN et al.[11] in their tables 2A and 2B as input, and specifying $\delta = 10\ nM^{-1}$, the program converged to a solution identical to theirs in 28 sec. The iteration time increases as an approximately linear function of $-\log\delta$. Although the iterative technique used in this algorithm is not very fast, it is quite adequate for laboratory use, and it is safe provided that the magnitudes of the concentrations of sites and ligands are not so disparate that the digit-carrying capacity of the calculator is exceeded. In our laboratory it has proved useful in simulating the nonspecific and the specific binding of progesterone and cortisol to the two classes of high-affinity sites thought to be present in rat uterine cytosol[14]. Its application to the validation of a filtration technique for the estimation of these high-affinity sites, a modification of the method of SANTI et al.[15], will be reported separately.

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