THE RECOMMENDED DIETARY ALLOWANCE FOR ASCORBIC ACID

Despite the potentially devastating consequences of the disease, scurvy is easily prevented. The recommended daily dietary allowance (RDA) for ascorbate was determined by the Food and Nutrition Board of the National Research Council on the basis of several estimates that are directly relevant to scurvy.[1] The human requirement is based on the amount necessary to cure or prevent scurvy while allowing for adequate reserves of ascorbate and on the amount of ascorbate metabolized daily.[1,57,65,70] In the United States the RDA is 60 mg per day for adults,[1] and in the United Kingdom it is less.

It is worthwhile to review some of the estimates on which the RDA is based. Clinical scurvy occurs when the body pool of ascorbic acid is less than 300 mg. It has been demonstrated in human volunteers that as little as 10 mg of ascorbic acid per day can cure or prevent scurvy. However, daily ingestion of only 10 mg of ascorbate for several weeks results in a body pool of ascorbate that is still not substantially above 300 mg.[57,66] Therefore, ascorbate metabolism, turnover rates, and the size of the body pool have been estimated in volunteers who have ingested radiolabeled ascorbate. When 77.5 mg of ascorbate is ingested daily, a pool of approximately 1500 mg can be maintained.[1,57,69,70] This amount of reserve can prevent clinical scurvy from developing for l to l½ months in volunteers given a scorbutic diet. Ascorbate catabolism has been estimated by giving subjects radiolabeled ascorbate immediately before scurvy was induced and measuring the urinary excretion of radiolabeled catabolic products. In these experiments, ascorbate catabolism was determined to be 2.2 to 4.1 percent of the existing body pool.[57,68-70] On the basis of these data, ascorbate catabolism was estimated to be 34 to 61.5 mg per day for a body pool of 1500 mg. It is important to remember, however, that these estimates were made in a study of subjects with scurvy rather than normal, healthy subjects.

In subjects with scurvy who were given ^{3}H-labeled or ^{14}C-labeled ascorbic acid, excretion of the radiolabel as ascorbic acid itself did not occur until the body pool of ascorbate was approximately 1500 mg.[68,69] Since constant dose of approximately 60 mg of ascorbate per day is necessary to create and maintain a body pool of 1500 mg, this daily intake will theoretically maintain the body pool, replenish catabolized ascorbate, and protect against scurvy for at least 30 days if ascorbate ingestion ceases abruptly. Thus, on the basis of all these estimates, the RDA for adults has been set at 60 mg per day.

IMPLICATIONS OF THE RDA FOR ASCORBIC ACID: PREVENTION OF SCURVY VERSUS OPTIMAL REQUIREMENTS

The simplest question to pose and yet the most difficult one to answer is whether the RDA for ascorbic acid is correct. The data clearly show that a daily intake of 60 mg will prevent scurvy with a margin of reserve; therefore, this allowance is clearly correct insofar as prevention of this deficiency disease is concerned. However, the question whether the amount of

ascorbic acid needed to prevent scurvy is equivalent to the optimal amount for human health remains unanswered.[2,65]

There is no clear relation between the biochemical roles of ascorbic acid and the deficiency disease scurvy.[16] Thus, the amount of ascorbate necessary to prevent scurvy may not be equivalent to the amount that satisfies diverse enzymatic needs for ascorbic acid. For example, in two studies of animals that did not have scurvy but were not receiving "optimal" amounts of dietary ascorbate, ascorbate-dependent enzyme function was still decreased.[31,40] Unfortunately, there have been few experiments in which various ascorbate contents of cells or animals have been rigorously correlated with enzyme activity.[38,39,62,71-73] In biochemical terms, there have been few demonstrations in cells or tissue of the apparent Michaelis constant or the apparent maximal reaction velocity of ascorbate using enzyme systems that in purified form depend on ascorbic acid for full activity.[32,39] Measurements of these and other biochemical determinants, such as cofactor transport into tissue, intracellular compartmentalization, and cofactor-enzyme accessibility, might reflect ascorbate needs in vivo more precisely than measurements indicative of scurvy.[38,39,74,79] However, it has not yet been established which biochemical function or functions would best mirror the optimal dietary level of ascorbate.[78]

The need for ascorbate can also be examined from a metabolic standpoint. Although a body pool of 1500 mg of ascorbic acid will prevent scurvy, this pool size may not be sufficient for optimal human health. Larger pools of ascorbate can be attained if intake is increased. Studies in male volunteers who ingested 200 mg of ascorbate daily (140 mg per day more than the RDA) have shown that the body pool can be expanded to 2300 to 2800 mg.[67] In addition, studies in rats and guinea pigs whose ascorbate dosage per kilogram of body weight exceeded the RDA have shown that it may be possible to expand the body pool even more markedly.[80-82] In human beings, however, it has been suggested that the ingestion of ascorbate in quantities sufficient to maintain a larger body pool is unwarranted.[1] The larger ascorbate pool is believed to be obtained at the expense of decreased gastrointestinal absorption and increased excretion of unmetabolized ascorbate. Yet, there is no evidence that these patterns of absorption and excretion are an indication of excessive ascorbate ingestion. Rather, they may be an unimportant consequence of maintaining higher body stores of ascorbate or of generating plasma-tissue gradients. Until the optimal size of the body pool of ascorbate is determined, these problems will remain unresolved.

Another approach to the problem of ascorbate requirements has been to study urinary excretion of ascorbic acid in humans. As discussed earlier, ascorbic acid does not appear to be excreted in the urine until the body pool of ascorbate reaches 1500 mg or more. However, these data may have no bearing on optimal ascorbate requirements, since there may be no relation between the threshold of ascorbate excretion and optimal tissue concentrations.

The requirement for ascorbate is also based in part on measurements of ascorbate catabolism, which generate estimates of daily ascorbate turnover. However, the most direct measurements of ascorbate catabolism have again been obtained in subjects who were depleted of ascorbic acid,[57,68,69] as opposed to the indirect values obtained with the use of pharmacokinetics.[70] In the depleted subjects, one would expect the catabolism of ascorbate to be minimal, given the limited supply. Indeed, catabolism seems to vary as a function of the amount of available ascorbate, as the Studies in animals discussed below indicate. Thus, it is not clear that the rate of ascorbic acid use would remain at 2.2 to 4.1 percent of body stores in subjects who were not ascorbate-poor. Measurements of ascorbate catabolism would be more meaningful if they were made in persons with an optimal body pool of ascorbate, but we do not yet know what the size of that optimal pool may be.

and are difficult to quantitate. Because we do not know what clinical or biochemical measure best reflects optimal ascorbate ingestion, the assays have been chosen empirically, and as a result, many of the data conflict. Furthermore, in animals as well as humans, it is worthwhile but very difficult to measure the additional effect of "stress," or perturbation of homeostasis. Although these issues are clearly important, they have not been subjected to intense critical investigation. In addition, the problem of optimal intake is sometimes approached emotionally, to the extent that scientific issues are not addressed.

Because no clear relation has been seen between scurvy and the biochemical behavior of ascorbate, there have been few attempts to use biochemistry to address the problem of ascorbate requirements. Yet, herein is the key to the problem. Ascorbate requirements must be examined both by studies in whole animals and by studies of ascorbate cell biology and biochemistry. The role of ascorbic acid in several enzyme systems is becoming known, and what remains to be studied is how those enzymes are regulated by ascorbic acid in cells, tissues, and animals. When the ways in which various enzymatic functions depend on ascorbic acid are characterized, these measurements can be brought to tissue and animal studies in a precise fashion with use of specific assays. In this way, variations in ascorbic acid concentrations can have measurable and meaningful biologic implications.

SOLVING THE PROBLEM OF THE **RDA** VERSUS THE OPTIMAL AMOUNT OF ASCORBATE

The data in animals suggest even more strongly than the data in humans that the RDA of ascorbate for the prevention of scurvy and the RDA for other measures of health are not similar. The resolution of this problem has been complicated by several factors. Although the assays for prevention of scurvy are straightforward, methods of determining optimal ascorbate ingestion are not. In animals as well as in human beings, many of these assays lack specificity

OPTIMAL DIETARY LEVELS: A SUMMARY

Our knowledge of the amount of cofactor required to prevent deficiency disease may or may not shed light on the problem of optimal dietary levels, which is a related but distinctly separate issue. I have proposed that the optimal dietary level of ascorbic acid may be diiferent from the RDA, and that it is important to determine this level. Although biochemical, enzymatic, and functional principles suggest that there are optimal ingestion levels for many cofactors, it is not entirely certain that there is such a level for ascorbate or for any other cofactor in human beings. It is also unclear whether optimal concentration is equivalent to tissue saturation. These issues can and will be resolved by biochemical and cellular studies in conjunction with investigations in animals.

Nearly half a century ago, the pioneers David Perla and Jesse Marmorston and their collaborators advanced one hypothesis concerning optimal intakes of the cofactor thiamine.[45] The fact that there have been no recent efforts to determine such intakes for thiamine or other cofactors does not mean that the issue should be left dormant. Indeed, the principles discussed above in connection with ascorbic acid can and should be applied to other vitamins. It is only through rigorous scientific investigation with open and unbiased minds that we will learn the optimal dietary levels of such cofactors as ascorbic acid. Knowledge of the optimal levels of cofactors will have profound implications for the practice of medicine and the health of ali of us.

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NEW CONCEPTS IN THE BIOLOGY AND BIOCHEMISTRY OF ASCORBIC ACID

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ASCORBIC acid, originally called vitamin C, is required for human health.[1] In human beings deprived of ascorbic acid, the deficiency disease scurvy develops and can be life threatening. Although a disease remarkably similar to scurvy was described by the ancient Egyptians,[2,3] it was not until 1753 that a Scottish physician, James Lind, systematically described scurvy and its prevention by dietary means.[4] Even then, the dietary requirements were controversial. For four decades the British navy refused to accept Lind's findings, and countless sailors continued to die unnecessarily from scurvy until lemon juice was finally included in sailors' rations.

Research since Lind's time has established that the dietary substances that prevent scurvy are those that contain ascorbic acid.[5,6] What has remained unclear is whether the amount of ascorbate necessary to prevent scurvy is similar to the amount necessary for optimal health. The issue of optimal dietary levels of ascorbic acid and other vitamins inevitably provokes controversy. In this article I will address the many issues that contribute to the debates about ascorbic acid and suggest new approaches to their resolution.

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Reprinted from the *New England Journal ofMedicine*
314:892-902 (April 3), 1986