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Reply

As pointed out in our earlier publications (*1-4*) we have shared for some time the concern of Drs. Baker, Frank, and Hutner regarding the nonspecificity of the competitive protein-binding assays (CPBAs) for vitamin B_{12} and folates. Indeed, we observed normal test results in patients who were clearly suffering from severe deficiencies of one or other of these essential nutrients. This fact, combined with the reality that the well-proven specific microbiologic growth assays are no longer available to the large majority of clinicians (because they are no longer acceptable to most clinical laboratories), led us to search for new, reliable, sensitive, and specific methods for assay of these and other essential nutrients.

We emphasize that our new radiometric microbiologic assays (RMAs) share nothing in common with these CPBAs; instead we have been able to utilize the selectivity and specificity of the microorganism, but have enhanced specificity, sensitivity, and precision of measurement by careful selection of the appropriate C-14-labeled substrate for each assay. As is detailed in our publications (*1,2,5*), we have reason to believe that coenzyme-dependent ("vitamin"-derived) pathways are being utilized in the presence of the biologically active forms of these essential nutrients, thus enhancing specificity. We believe that we are combining the unique properties of these microorganisms with the sensitivity and precision of measurement of a physical event (i.e., radioactive decay).

In addition, our required "growth" period is only *18 hr* and the measurements of many assay vials are fully automated. Turbid solutions, precipitates, or other debris do not interfere with our RMAs, which are thus readily applicable to previously much more tedious assays, such as that of foodstuffs (*6*).

It certainly was not our intent to offend the authors of the single most definitive publication on the use of *T. pyriformis* for the assay of nicotinic acid (*7*). On the other hand, we found the data as presented in that paper difficult to assess, since no "N", means, or standard deviations are given for any of their measurements. Tables 2 and 3 in that paper indicate a growth response to certain amounts of nicotinuric acid, such that if these were present in whole blood, this biologically inert substance could clearly mask a deficiency state of "niacin" in man.

We do agree that trigonelline does not promote the growth of *T. pyriformis;* however, other compounds such as nicotinmethylamide and nicotinethylamide (although biologically active in dogs (*8*)), do not have the full potency of nicotinic acid and its amide in promoting the growth of *T. pyriformis*. We do know that *L. plantarum* responds to nicotinuric acid (measured turbidimetrically) as pointed out in our paper (*5*); however, we have also indicated that this nonspecificity was largely circumvented when the compound was measured using our RMA.

Our earlier detailed review of available literature on pellagra indicated that all studies, regardless of the methods then available, showed a great overlap of values between patients with pellagra and normal subjects. It has recently been noted that we continue to see this disease in our own medical institutions (*9*) and that the often long-delayed proper diagnosis of the described patients almost certainly represented only the "tip of the iceberg." It was these facts that led us, after development of RMAs for vitamin B_{12} and folate, to apply this entirely new combination of standard technologies and readily available equipment to the assay of "niacin," and then to test it carefully for specificity. For the initiate to this field, we should point out that any equipment for detecting β radiation (from a simple ionization chamber to an automated liquid-scintillation counter) can be adapted for use with our RMAs (*10-12*). Certainly, the automated equipment used by us has only recently been spreading to clinical laboratories. On the other hand, we share the concept of "regionalization" for infrequently needed tests and/or those requiring special expertise and equipment.

We wish to thank the authors of the above letter for sharing their concerns with the readers of the *Journal of Nuclear Medicine*. We hope they will agree with us that ultimately it will be these readers who will make the choice of the method most suited to their needs.

PATRICIA A. McINTYRE
TOMAS R. GUILARTE
JUDITH A. KERTCHER
MARIANNE F. CHEN
AGATHA A. RIDER
Johns Hopkins Medical Institutions
Baltimore, Maryland

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