Riboflavin deficiency in women taking oral contraceptive agents[1, 2]

Leonard J. Newman, M.D., Rafael Lopez, M.D., Harold S. Cole, M.D., Maria C. Boria, M.D., and Jack M. Cooperman, Ph.D.

ABSTRACT The effect of oral contraceptive agents (OCA) on riboflavin nutritional status of women of child-bearing age in a low socioeconomic population was studied. For a control group, 100 women in the same age and socioeconomic group using alternate forms of contraception were selected. Riboflavin deficiency was determined by measuring erythrocyte glutathione reductase activity, a reliable index of the deficiency. None of the women was on vitamin supplements or had clinical conditions effecting dietary intake or utilization. Eleven of 100 women in the control group had biochemical evidence of deficiency. This compared to 24 of 56 OCA users who were deficient. The frequency of deficiency increased among those on OCA for longer periods of time. Thirteen of 17 OCA users for 3 years or more, compared to 11 of 39 users under 3 years were deficient. There were no discernable dietary differences between the groups. These studies demonstrate that riboflavin deficiency is a problem of women in the lower socioeconomic level in the child-bearing age. The use of OCA aggravates the prevalence of deficiency. *Am. J. Clin. Nutr.* 31: 247-249, 1978.

The effects of oral contraceptive agents (OCA) on vitamin nutrition have been reviewed (1-3). However, few studies on riboflavin deficiency in OCA users were listed. Recently, a study in Thailand (4) demonstrated that the activity of erythrocyte glutathione reductase (EGR), a flavin adenine dinucleotide (FAD) containing enzyme, in women on OCA was lower than in a control group. Briggs and Briggs (5) reported low urinary riboflavin excretion in women on OCA in Zambia. We wish to report our studies of riboflavin deficiency in women of low socioeconomic status using oral contraceptives for varying periods of time.

Materials and methods

The study was performed in the family planning clinics of a large municipal hospital in East Harlem, New York City, a predominantly black and Hispanic-American neighborhood. A control group of 100 females, 17 to 35 years old who were on contraceptive devices other than OCA or on no form of contraception was selected. Specifically excluded were those who gave birth within 1 year of the study, those on vitamin supplements, and those with malabsorption or metabolic diseases effecting nutriture.

The study group consisted of 56 women 18 to 35 years old and on OCA for intervals ranging from 1 month to 13 years. As in the controls, those with underlying illness or on vitamin supplements were excluded.

Dietary recalls revealed similar dietary regimens in both groups.

Five milliliters of venous blood were collected from each subject in tubes containing 1.5 ml of ACD solution (7.3 g anhydrous citric acid, 22 g sodium citrate $\cdot 2\ H_2O$ and 24.5 g dextrose $\cdot H_2O$ per liter). The samples were refrigerated and the erythrocytes separated by centrifugation within 4 hrs of collection. EGR activity of the hemolyzate was determined by a modification of the method of Glatzle et al. (6) as previously described in detail (7). The assay is run both in the presence and absence of added FAD, and the end point is the decrease in optical density (OD) units at 340 nm owing to the oxidation of NADPH. The results are expressed as the activity coefficient (AC) which is the ratio of differences in OD units with added FAD to that without FAD and is a measure of the degree of saturation of the apoenzyme with coenzyme. Normal values range from 0.9 to 1.2 (6, 7).

The oral contraceptives agent were either of two preparations, 1 mg norethinodrone and 50 μg mestranol (Norinyl 1/50) or 0.5 mg norgestrel and 50 μg ethinyl estradiol (Ovral).

[1]From the Section of Nutrition of the Department of Community and Preventive Medicine, the Department of Pediatrics, and the Department of Obstetrics and Gynecology, New York Medical College, New York, New York 10029.

[2]Address reprint requests to: Leonard J. Newman, M.D., Department of Pediatrics, New York Medical College, 1249 Fifth Avenue, New York, New York 10029.

Results

In the control group 11 of 100 women had abnormally elevated AC values indicative of riboflavin deficiency. Twenty-four of 56 women on OCA had AC values above 1.2, the upper limit of normalcy. The difference between the two groups were statistically significant ($P < 0.01$ by the χ-square test). The data in Figure 1 show that the deficiency was more frequent in the women on OCA for longer periods of time. Statistical analysis revealed that the prevalence of deficiency was significantly greater than in the controls both for the women on OCA for periods of 1 month to 2 years (nine of 24, $P < 0.05$) and those on OCA for periods of 37 months to 13 years (15 of 23, $P < 0.01$).

In none of the women with elevated AC values were there any clinical signs of riboflavin deficiency. There was no correlation of deficiency with the type of OCA used.

Discussion

These studies revealed that riboflavin deficiency is not an uncommon occurrence in a group of women of child-bearing age and of a low socioeconomic level. This may be owing to the fact that riboflavin naturally occurs in relatively expensive foods such as milk, eggs, meat, and green leafy vegetables and is low in cereal grains and legumes. The latter are the staple foodstuffs for many of these subjects. Among Puerto Ricans it has been shown that their daily regimen is low in riboflavin (8).

Among the users of OCA, the prevalence of riboflavin deficiency was markedly higher than in the control group. From dietary histories there were no discernable difference in vitamin intake between these groups. This would tend to implicate the OCA as exerting an effect on riboflavin metabolism.

The prevalency of riboflavin deficiency was greatest among those on OCA for longest periods of time. That this was not solely related to the age of the subjects can be adduced from the observation that in the controls the prevalence of deficiency did not correlate with age. It may be that women on OCA from 1 to 6 months may have been deficient before initiating this mode of contraception.

These results are in agreement with those reported from Thailand (4), Zambia (5), and India (9) where the increased prevalence of riboflavin deficiency found among

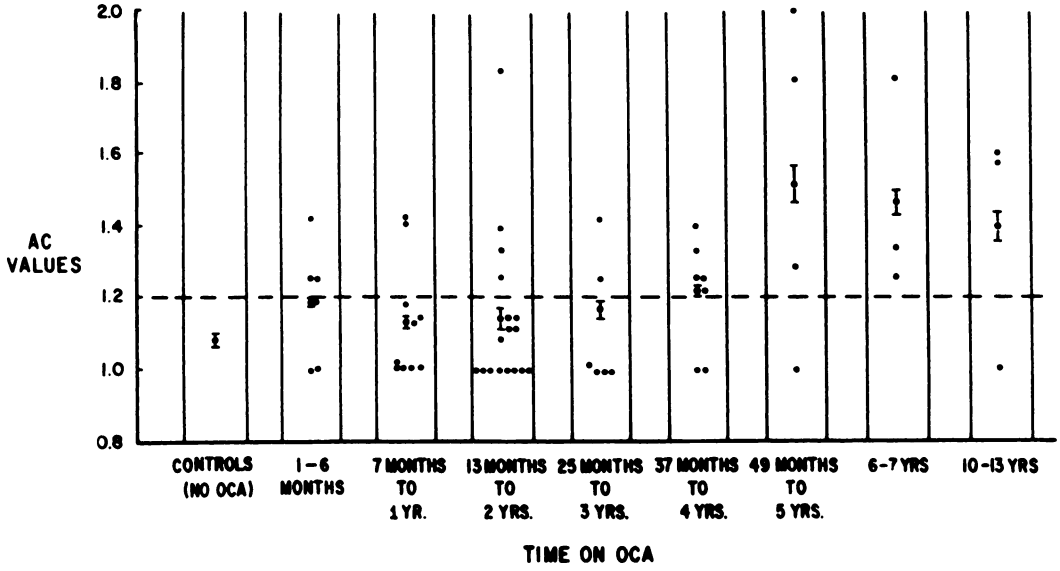

FIG. 1. Prevalence of riboflavin deficiency in OCA users related to duration of use. AC values above 1.2 indicate riboflavin deficiency. In each time group, individual values are plotted with the mean and standard error given.

OCA users could not be attributed to dietary intake of this vitamin. Wynn (2), pointed out that the occurrence of riboflavin deficiency may be harmful in women with glucose-6-phosphate dehydrogenase deficiency. In such subjects, the EGR levels are elevated and this has been postulated to compensate for the metabolic derangement in the erythrocytes. A reduction in EGR may nullify this mechanism. This may have important clinical implications for blacks with glucose-6-phosphate deficiency.

In experiments with riboflavin deficient pregnant rats, Warkany and coworkers (10, 11) demonstrated a host of anomalies in the offspring. In addition infant mortality was high. Early studies could detect no correlation between riboflavin deficiency and the occurrence of fetal anomalies in the human (12). However, the method used to detect riboflavin deficiency was relatively nonspecific.

A recent study of riboflavin deficiency in human pregnancy using the EGR test revealed that 25% of women in the first trimester and increasing to 40% at term were deficient (13). However, the authors do not state whether any of these women were on OCA previous to conception. With the wide use of OCA it is possible that riboflavin deficiency will occur with frequency in pregnant women who were previous users of OCA, and the effect of this on the developing fetus is still to be determined.

It has been postulated that the effect of OCA on riboflavin metabolism may be on interference with gastrointestinal absorption, metabolic conversion to active coenzyme forms or on binding of the coenzyme to the apoenzyme (2). Further studies are required to determine which component of OCA is responsible for its effect.

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