indole uptake and indole glycerol phosphate formation differ significantly.

Results similar to this have been observed in experiments on indole uptake with mycelial pads of strain td_{2} (13), suggesting that several products may be formed from indole during the course of the reaction. It should be emphasized however, that close agreement is obtained between indole uptake and the number of units of CRM in the extract, whether different aliquots of the same preparation or different preparations are compared. Furthermore, extracts of mutant strain td1, a mutant which forms neither tryptophan synthetase nor CRM (1, 4, 5, 14), exhibit no "indole uptake" activity.

Rabbit anti-td₂ CRM sera (5) were tested for their ability to inhibit the indole reaction catalyzed by td2-CRMcontaining preparations. It was found that unabsorbed anti-td₂ CRM serum, and anti-td₂ CRM serum absorbed with extracts of strain td_1 , a mutant which lacks both tryptophan synthetase and CRM (5) completely neutralize enzyme activity. As is shown in Table 1, anti-td₁ serum (5) has no inhibitory effect.

These results seem to support the proposal that the CRM protein in Neurospora mutant td₂ is the protein responsible for catalyzing one or more reactions involving indole. It would appear most likely that td₂-CRM represents one of a variety of possible types of genetically altered tryptophan synthetase molecules. In this case an alteration in protein structure seems to have eliminated the reactivity of the protein with L-serine, albeit an indole-combining site is still retained. It is well known that specific suppressor mutations are capable of partially restoring tryptophan synthetase activity in certain CRM-forming td mutants of *Neurospora* (1). Perhaps one mechanism of action of such suppressor genes involves the conversion of a CRM protein to active tryptophan synthetase by reactivating an inactive catalytic site on the CRM protein (7, 15).

> SIGMUND R. SUSKIND ELKE JORDAN

McCollum-Pratt Institute, Johns Hopkins University, Baltimore, Maryland

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Fat Changes during Adolescence

Abstract. Lower thoracic fat, as measured on serial chest plates of 259 children in Ohio, increased in girls, between ages 6.5 and 14.5 years and, in boys, between ages 6.5 and 11.5 years. No evidence for a marked loss of "baby" fat in adolescence or for "waves" of fattening around the time of puberty was found.

The literature on human growth refers to a number of changes in body fat during adolescence, including a loss in "baby" fat that is popularly believed to occur and the existence of several waves of fattening that supposedly occur either before or after the spurt in stature (1). However, currently available data are insufficient to confirm these generalizations, and the still-limited findings are contradictory.

In the present study, outer fat was investigated on the lower thorax (2) at the level of the tenth rib, in 259 regular participants in the Fels longitudinal program, by means of serial posteroanterior chest radiographs. Sex-specific samples ranged from 16 to 93, the age range was 6.5 to 17.5 years, and the average sample for each age group was 124. In the treatment of the data, median values were used because of the marked skewness of all 24 distributions.

As is shown in Fig. 1, there was a parallel increase in fat on the lower thorax in both sexes from the 6th through the 11th year, the median values for the girls being about 1 mm (or 40 percent) above those for the boys. Thereafter, fat on the lower thorax continued to increase in the girls, reaching a thickness of 8 to 9 mm by the 14th year, while in the boys it stabilized at about 4.5 mm between the 11th and the 17th years. No loss of "baby" fat was evident for the girls in this cross-sectional analysis, and the tiny (0.2 -mm)decrease in the median values for the boys between 11 and 13 years was not statistically significant.

Further analysis of the data, on a longitudinal basis and on the basis of physiological events rather than chronological

age, yielded similar results. For the girls, rearrangement of the fat measurements relative to the individual age at menarche or to the year of the maximum spurt in stature confirmed the observation that there is a fairly uniform increase in fat from childhood on. In the boys, the maximum accumulation of fat on the lower thorax generally preceded the year of the spurt in stature, and there was a suggestion that there is a small decrease about 2 years after this event.

The lack of data that would indicate a period of fat loss, or of "waves" of fattening in girls, was in agreement with earlier findings on the same population sample (3) and with data on fat in the calves of girls in Boston (4). The trends in fat on the lower thorax of boys in Ohio, while in general agreement with trends in fat at other sites in the same group (3), were quite different from trends in fat in the calves of Boston boys (4) and different from trends found by pinch-caliper measurements of boys in California (1). While increase in fat in the boys was noted in all three studies, there was no agreement about when it began, when it terminated, when there was a fat loss, or when there was a steady-state leveling off.

These discrepancies may be due to differences in the sites selected, since fat may increase on one part of the body and decrease simultaneously on another part during growth (5). Alternatively, the divergent trends apparently exhibited by California, Ohio, and Massachu-



Fig. 1. Continuous increase in lower thoracic fat in girls and parallel increase in boys, terminating at 11.5 years. By 14.5 years the adult female/male fat ratio of 180 percent (2) has been attained.

setts boys may reflect true differences in the populations sampled. The simplest explanation that fits our data is that of a steady increase in outer fat in both sexes, with a temporary interruption in the male during the period of steroid hormone differentiation.

STANLEY M. GARN

JOAN A. HASKELL

Physical Growth Department, Fels Research Institute. Yellow Springs, Ohio

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Dissolution of Interlayers from Intergradient Soil Clays after Preheating at 400°C

Abstract. Dehydroxylated interlayers have been removed from chlorite-vermiculite-montmorillonite intergrades by boiling the preheated sample in 0.5N NaOH for 2.5 minutes. Elements extracted can be conveniently analyzed spectrophotometrically. A convenient method of estimating the amount of elements present in interlayer positions and preparing the sample for analysis for expanding and nonexpanding mineral components is thus provided.

An intergradient chlorite-vermiculitemontmorillonite is one of a series of layer silicates having heat stability of the 14-A spacing, cation exchange capacity, and specific surface intermediate between those of the nonexpanding mineral chlorite and the two expansible and collapsible minerals, vermiculite and montmorillonite. Layer silicates with properties intergradient between those of chlorite and expanding layer silicates have been identified in several soils in widely separated areas of the United States (1, 2) and in England (3)

The intergradient properties of such clays, relative to properties of the typical minerals, have confounded qualitative identification and prevented quantitative determination of each of the component clay minerals in soils. Also, the aluminum generally present in intergradient clays of soils actively affects chemical reactions, particularly those of soil acidity and liming, phosphorus fixation, and potassium fixation.

It has been found that the elements which give the intergradient properties can be dissolved from between the layers after dehydroxylation of the interlayers of preheating the dry, loose, hydrogen-saturated clay at 400°C for 4 hours. The dissolution is accomplished by boiling the preheated clay sample in 0.5N NaOH (at a ratio of 40 ml of NaOH to 40 mg of clay) for 21/2 minutes, which is the differential dissolution procedure of Hashimoto and Jackson (4). The Al and Si dissolved by the treatment are immediately determined colorimetrically (5). The iron oxide precipitated during the NaOH treatment is removed by the dithionite-citrate-bicarbonate method (6; 7, p. 57). Removal of allophane from the clay by the same dissolution procedure applied to a sample with only 110°C preheating (4) permits calculation of the Al that is made soluble by the $400\,^{\circ}\mathrm{C}$ preheating.

Elements dissolved from the samples preheated at 110° and 300°C had a small effect on the intergradient properties, as is shown by some increase in intensity of the 18-A peak (Fig. 1) at the expense of the 14-A peak. However, the NaOH treatment of a different sample preheated at 400°C resulted in a marked increase in the amount of material which expanded to 18-A with Mg saturation and glycerol solvation and in the amount which collapsed to 10-A on K saturation and heating at 300°C in x-ray diffraction preparations (7, p. 184).

A small amount (about 0.6 percent) more alumina removed after preheating at 400°C than after preheating at 300°C is responsible for the marked difference in the 14-to-10-A collapse in the x-ray preparation (Fig. 1). This aluminous interlayer which impaired 14-to-10-A collapse on heating at 300°C is stable to higher temperatures than is free Al(OH)₃ as gibbsite, but to lower temperatures than complete $Al(OH)_3$ interlayers in chlorite. This is interpreted to indicate the presence in the intergrade of islands of positively charged hydroxy aluminum which bridge between potentially collapsible (and expansible) silicate layers. The percentage of alumina removed, when compared with that in complete gibbsite-like interlayers in aluminous chlorite, indicates that the islands in the intergrade are of limited extent.

Preheating the clay at 500°C destroyed the kaolinite, as is shown by the loss of the 7.2- and 3.57-A x-ray diffraction peaks (Fig. 1). The possibility that the 7.2-A peak is indicative of chlorite is ruled out by the absence of 14- and 4.6-4.8-A peaks (8) in the 400°C/K/ 300°C sample (Fig. 1) and by the fact that soil chlorites which lose their 7-A peak and reinforce their 14-A peak on



Fig. 1. Elements extracted and x-ray diffraction curves before and after NaOH extraction of the 1–0.2 μ fraction of Crosby soil, A₂ horizon, with various temperatures of preheating, $f(t^{\circ}C)$, K or Mg saturation, and glycerol solvation followed by 25° or 300°C heating of slides for x-ray diffraction.

500°C preheating are not appreciably dissolved by subsequent NaOH boiling (9). The differences (Fig. 1) between the alumina and silica extracted by the same NaOH extraction procedure (4) after the 400° and 500°C preheatings allocate to 15.1 and 15.5 percent kaolinite based on the two elements, respectively. Considerable increase in intensity of the 10-A peak (Fig. 1, 500°C/K/ $300^{\circ}C$ versus $400^{\circ}C/K/300^{\circ}C$) may be due to increase in ordering produced by the removal of a small amount more interlayer aluminum after preheating at 500°C than was removed after preheating at 400°C. Thus the alumina allocated to kaolinite may be slightly high despite the agreement between the allocations to kaolinite of alumina and silica. A little excess silica must be allocated to a small amount of nontronite destroyed, as is shown by the small increase in iron oxides released by 500°C preheating over those released by 400°C preheating; this would allow for a small amount more alumina from the intergrade made soluble by the 500°C preheating.

The dehydroxylation-NaOH method of dissolving interlayers from layer silicate intergrades was found to be more effective, more rapid, and far more convenient for analysis of the elements extracted than the citrate (2) or fluoride (10) methods previously available. This method was tested on intergradient clays from Tatum soil (Virginia) and Cookeville soil (Kentucky) and was found to give results comparable to the above results with the Crosby soil clay (11).

J. B. DIXON M. L. JACKSON

Department of Soils, University of Wisconsin, Madison

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