in the mitochondria. The microsomal content is much lower and is only about 5% of the total cellular folic acid. In view of the known functions of folic acid, the higher content of folic acid in mitochondria is striking. This is further corroborated by the higher uptake of radioactive folate by mitochondria (Table II). The ratio of mitochondrial to microsomal folic-2-C^{14} acid content in these uptake experiments varies from about 5–8. The same ratio in the normal mouse is approximately 3.5. SELIG and SANKAR[6] found that addition of folate enhanced the mouse liver mitochondrial oxidation of pyruvate in the presence of NAD. This leads us to wonder whether mitochondrial folate may not be involved in electron transport or in its regulation. In view of the fact that dihydrofolic reductase may use NADP and/or NAD depending on the pH of the reaction medium[7], it is conceivable that folate may be involved in the reactions that govern the NADPH/NAD ratios in mouse liver. Many folate-linked enzymes have higher activity in cytoplasm[8] than in the mitochondria. This adds support to the concept of a role for folate in mitochondrial electron transport rather than in mitochondrial folate-linked enzymes. Further studies to answer this question are in progress.

Table II. Percentage distribution of radioactive folate in mouse liver subcellular fractions

Cell fraction	% Distribution at time elapsed after administration of folate-2-^{14}C	
	30 min	120 min
Nuclei	19.5	14.9
Mitochondria	14.3	26.0
Microsomes	1.6	4.6
Cell sap	64.6	54.5

Zusammenfassung. Eine bedeutende Menge der gesamten Folsäure der Mäuseleberzellen ist in den Mitochondrien vorhanden. Auch nach Inkubation des Leberhomogenats mit radioaktiver Folsäure wird der Grossteil der Radioaktivität im mitochondrialen Anteil gefunden.

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Changes in Free Sugar During the Germination of Pea Seeds

Among the free oligosaccharides, sucrose, raffinose, and stachyose have been found in seeds of Leguminosae[1], Cruciferae[2], and Liliaceae[3]. A large amount of stachyose was found in peas[4], and the distribution of these oligosaccharides of the raffinose family in peas and lima beans during the course of ripening periods has been studied[5,6]. In experiments with pea extracts, it has been suggested that the sucrose is synthetized from UDPG and D-fructose[7,8]. In analogy with the formation of sucrose in plants, D-galactose-1-phosphate or D-galactose nucleotides have been suggested as intermediates in the formation of raffinose and stachyose from sucrose by enzymatic systems carrying on transgalactosylation reactions[9]. In the course of a study on germination of soybeans, α-galactosidase which is responsible for the splitting of D-galactose from the reserve oligosaccharides of the raffinose family, and enzymes which participate in the metabolsim of D-galactose were found[10].

In the study described here, individual free sugars were analyzed during the course of the germination of pea seeds and we observed the reverse of the process by which the reserve oligosaccharides of the raffinose family are formed during the ripening process.

Dry pea seeds (*Pisum sativum*, variety Early Perfection) produced at this Station were selected and soaked in water containing 10% (v/v) of Clorox for 18 h at room temperature. The rehydrated seeds were placed in petri dishes to germinate at room temperature (23 °C). Germinated peas were taken at intervals from 0 to 100 h and extracted with 80% alcohol for the analysis of individual free sugars by a quantitative paper chromatographic method[5,11].

It was observed (Figure) that the decrease in the absolute amount of stachyose and raffinose is accompanied by a corresponding increase in sucrose during germination of pea seeds. The amount of the only reducing hexose (glucose) monitored held constant at 0.5–1.0 mg/grain. Fructose and galactose were observed in trace amounts only.

Apparently α-galactosidase and galactokinase systems were very active at the early stages of germination of pea seeds and reversed the transgalactosylation reaction of the ripening process of peas. Therefore, stachyose and raffinose were hydrolyzed to galactose and sucrose, and the liberated galactose was converted rapidly into the

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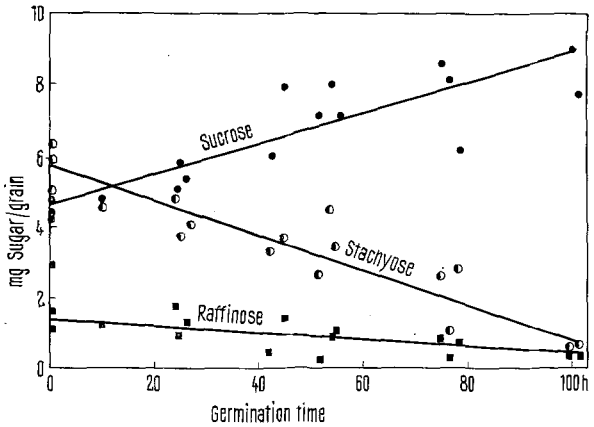
other metabolites. It is interesting to note that only galactose liberated from the reserve oligosaccharides actively participated in the metabolic processes of germinating pea seeds, while sucrose did not. On the other hand, sucrose is reported to be utilized rapidly during germination process of soybeans[10].

These results indicate that α-galactosidase is the major active enzyme system in pea seeds in the process of utilization of reserve oligosaccharides at the early stage of germination, and, therefore, it caused the oligosaccharides of the raffinose family to decrease in concentration, and free sucrose concentration is to increase.



Changes in concentrations of pea seed oligosaccharides during germination.

Zusammenfassung. Bei biochemischen Untersuchungen während der Keimung von *Pisum sativum* wurden Veränderungen im Gehalt von freien Zuckern beobachtet. α-Galaktosidase ist das aktivste Enzym unter den verschiedenen Enzymsystemen der Erbsen, die im Anfangsstadium der Keimung Reservezucker verwerten, wobei die Konzentration der Oligosaccharide abnimmt, während diejenige von Saccharose eine Steigerung erfährt.

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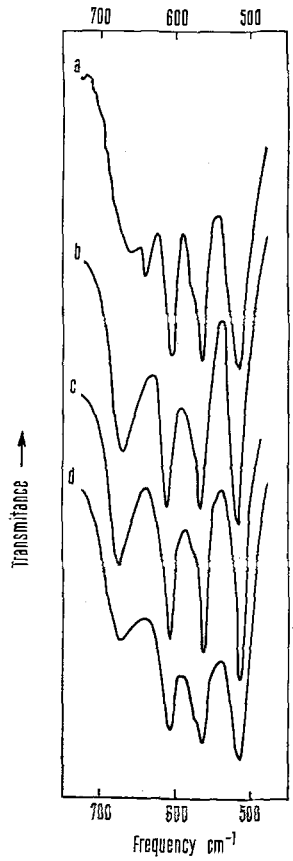
On the Nature of Calcium Phosphates in Urinary Calculi

Among the substances occurring most commonly in urinary calculi, calcium phosphates are, along with calcium oxalates, the ones found most frequently (thus, in our recent study of some 120 human uroliths from Macedonia (Yugoslavia)[1], we have found calcium phosphates in almost half of the investigated stones). Despite this fact, however, the true chemical nature of the calcium phosphate constituents is difficult to determine, mainly because they appear to be poorly crystallized and are, moreover, found almost always in an intimate mixture with calcium oxalates (apparently as a result of epitaxial overgrowth[2]). Having a long-standing interest in the IR-spectroscopy of calcium phosphates[3–5], we employed this technique in an attempt to solve, at least partly, this interesting problem.

Materials and method. The IR-spectra (recorded of KBr pressed discs on a Perkin-Elmer 521 Infrared Spectrophotometer) of the calculi were compared with the spectra (recorded by us or published) of the common stone-forming compounds. Artificial mixtures of some calcium phosphates (carbonatoapatite, hydroxyapatite, octacalcium phosphate) with calcium oxalates were prepared and their spectra also recorded. Particular attention was paid to the 750–500 cm^{-1} region in which the absorption bands are relatively sharp (cf. Figure) and their frequency can thus be measured with a rather high accuracy. Qualitative chemical tests were also performed.

Results and discussion. The analysis of the spectra showed that the calcium phosphate constituent of the



IR-spectra of (a) an artificial hydroxyapatite – Ca oxalate mixture; (b) an artificial carbonate-apatite – Ca oxalate mixture; (c) an artificial octacalcium phosphate – Ca oxalate mixture; (d) a calculus which does *not* contain carbonate-apatite.

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