dark period (12 h). Maximum light intensity, at the plants was 1300 f. c. The plants used had pods 12 days past anthesis. The first bloom node leaf of each of four plants was inserted into a glass manifold chamber and sealed with plastic clay. The pressure in the chamber was reduced to 80 mm mercury after which it slowly returned to atmospheric pressure. Radioactive carbon dioxide ($C^{14}O_2$) was generated from $BaC^{14}O_3$ and allowed to enter the sealed manifold chamber. 150 μc of C^{14} was supplied to the chamber. The leaves were allowed to photosynthesize in light at 850 f. c. for 3 h. At the end of this period the contents of the manifold were flushed into a 20% KOH solution.

The plants were divided into six parts: the leaf to which the $C^{14}O_2$ was supplied ('supply leaf'); the stem and leaves above the first bloom node ('stem above'); the stem and leaves below the first bloom node ('stem below'), the roots; the carpel; and the ovules. The plant parts were harvested, frozen and macerated in liquid nitrogen, and the powdered material dried for 24 h at 60°C. The dried plant material was extracted with 80% ethanol and filtered through Whatman no. 1 filter paper. The resulting extracts were brought to a volume of 50 ml, and 2 ml aliquots were removed, taken to dryness in 1-inch planchets, and counted in a proportional counter.

Results and Discussion. Carbon-14 activity in the plant parts is given in Table I. About 90% of the C^{14} activity was localized in the pod (carpel plus ovules). Relatively little (1%) of the C^{14}-labeled compounds was found in the roots and 9% of the transported carbon-14 was found in the stem and the leaves (St_A and St_B). Aliquots of the 80% ethanol extracts from the 'supply leaf', from the carpel, and from the ovules of individual plants were spotted on Whatman no 1 filter paper and subjected to descending chromatography in a *n*-butanol:ethanol (95%): water solvent 52:32:16. Following 40 h of development at a temperature of about 75°F the papers were sprayed with *p*-anisidine-PO_4 and developed at 95°F[3]. Strips ($1^1/_2$ in. wide) from these chromatograms were analyzed with a strip scanner attached to a gas flow Geiger counter with a micromil window (150 mg/cm²). The distance from the origin to the spots developed with *p*-anisidine-PO_4 and the distance from the origin to the zones of radioactivity was measured (Table II). Unlabeled sucrose, fructose, and glucose were tentatively identified in the carpel by their color development and sucrose was found to be located at the same distance (D_2) as the 'known' sucrose from its radioactivity. Radioactive sucrose was also found to be present in the ovules and in the 'supply leaf' following the 3 h period. With the aliquot used, only radioactive sucrose was found in these two plant parts. An aliquot of the extract from the carpel was treated with a 1% solution of invertase, then two-dimensionally cochromatographed with glucose and fructose using *n*-butanol:ethanol (95%):water (52:32:16) and phenol:water (4:1) as the solvent systems.

Only radioactivity in the glucose and fructose 'positions' was found. These results suggest that sucrose is the principal form of sugar accumulated in the leaf and may also be the form in which the photosynthate is translocated to the pod in *Pisum sativum* during the 3 h period. This finding is in agreement with that found for grape[4], and for soybean[5]. The similarity of the distribution pattern for labeled photosynthate and phosphorus-32[2] suggests that the controlling mechanism relating the 'loading' of compounds into the phloem in the leaf and the translocation to sites of utilization in remote organs may be partially similar.

Tab. I. The distribution of 80% ethanol soluble carbon-14 labeled compounds in different parts of *Pisum sativum*

Plant part	CPM × 10^3[a]	$S\bar{x}$%	% C^{14} translocated
Carpel	546	± 28	31 | —90 (pod)
Ovules	1031	± 4	59 |
Stem above	68	± 38	4
Stem below	83	± 38	5
Roots	14	± 26	1
Total	1742	± 4	—
Applied leaf	7879	± 8	—

[a] Each value is the average of the counts made from the respective part of 4 plants.

$$S\bar{x}\% = \frac{S \times 100}{\sqrt{n} \times \text{Avg. CPM}}$$

Tab. II. The identification of sugars in extracts of *Pisum sativum*

Compound	D_1[a]	Leaf D_2	Extract Carpel D_2	Ovules D_2
Sucrose	9.9	9.9	9.3	10.0
Fructose	15.5	0	0	0
Glucose	19.0	0	0	0

[a] D_1 = distance in inches of the spot from the origin developed with *p*-anisidine-PO_4. D_2 = distance in inches of the spot from origin located by measurement of radioactivity.

Zusammenfassung. Es wurde eine starke Verschiebung des C^{14}-Anteils des Achselblattes von *Pisum sativum* zur Frucht hin, die in der Achsel dieses Blattes entspringt, beobachtet. Bis zu 90% des C^{14} (80% Alkoholfraktion) der Pflanze befand sich nach 3 h in der Frucht. Der Hauptanteil des C^{14} dieses Blattes, der Hülse und der Samenanlagen wurde im Rohrzucker festgestellt.

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Poliovirus Production in HeLa Cells in Presence of Colchicine

Previously the effect of colchicine on poliovirus production was investigated in Maitland cultures of Rhesus kidney[1]). The behaviour of hydrolytic enzymes was also investigated[1], but not the cytological changes. In this preliminary report we describe biological findings on human carcinom a_1 cells (HeLa) treated with large dose of the antimitotic. The cells were subcultured with complete medium[2], in roller tubes, or coverslip cultures.

[1] E. KOVÁCS, Exper. *14*, 295 (1958).

2-days explants were inoculated with 0.5 ml undiluted Type I poliovirus (Mahoney strain) exhibiting a titer of 8.6 ID_{50}/ml. Direct adsorption of the virus was allowed for 30 min. The unadsorbed inoculum was decanted, the cells were washed 3 times with physiological salt solution (PBS) and incubated with complete medium, containing 1.5 µg colchicine / 1.5 ml. Controls were inoculated with pseudo-inoculum containing no virus[3]. 40 tube cultures were used for an experiment, 20 were inoculated with virus, 20 served as uninoculated controls, without colchicine, to compare virus production in unmedicated HeLa cells. Coverslip cultures were processed by vital staining, or the usual histological techniques at various time intervals. The cytological findings will be reported separately. The supernatant media of the infected cell cultures, exhibiting 3 to 4 + cytopathic effect, were pooled 24 h after infection. Titration of the two infected groups was carried out on tube cultures of HeLa cells. The virus production is illustrated in the Table.

The results of two assays are tabulated for each group. Both the colchicine treated and untreated systems produced large amount of poliovirus. The titers, in contrast to previous experiments in Maitland cultures of Rhesus kidney cortex[1], were high. There was about the *same* or even higher virus growth in the presence of colchicine as in untreated cultures. The difference, however, was within the error of the titration techniques used.

Effect of colchicine on poliovirus production in tissue culture

Materials	Repeated titrations[a]	Virus growth[b] in presence of colchicine	Virus growth[b] in absence of colchicine
Pooled supernatant of 10 HeLa cell tube cultures	Assay 1	7.60	7.75
Same as above	Assay 2	8.25	8.20

[a] On tube cultures of HeLa cells. [b] Titers as negat. logarithms ID_{50}/ml.

The findings described are of some importance. They suggest that the biochemical set-up of the mitosis may be selectively inhibited, without interferring with the synthesis of an RNA virus. Especially the spindle formation, the point of attack of colchicine, is fully independent from processes leading to the propagation of poliovirus. Thus not all the physiological functions of a cell are necessary for the biosynthesis of a virus, but there is certain *selectivity* in this phenomenon.

Systematic exploration of the behaviour of DNA-containing viruses in regard to colchicine treatment is planned, together with pertinent biochemical studies.

Zusammenfassung. Die Vermehrung des Poliovirus in HeLa Zellen war in Gegenwart oder Abwesenheit starker Colchicindosen *gleich hoch.* Die biochemischen Prozesse bei der Spindelbildung sind scheinbar unabhängig von denen der Biosynthese eines RNA-Virus.

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Aktivitätsänderungen der Lactatdehydrogenase im Mäuseplasma durch Beimpfung mit virushaltigen Geschwulstfiltraten oder virushaltigen Gewebekulturen[1]

Durch tierexperimentelle und elektronenmikroskopische Untersuchungen wurde gesichert, dass in einigen Impfgeschwülsten der Maus virusartige Körper enthalten sind, die bei neugeborenen Mäusen myeloische Leukämien verursachen[2,3]. Während der monatelangen Latenzzeit bis zur Entwicklung der Leukämien gibt es praktisch keine Nachweismöglichkeiten des Virus oder seiner Infektionsfolgen. In diesem Zusammenhang sind Untersuchungen von Bedeutung, in denen auffallende Erhöhungen der Fermentaktivität von Lactatdehydrogenase (LDH) nach Injektion von Geschwulstfiltraten berichtet wird[4]; die Autoren wurden durch ihre Ergebnisse veranlasst, ein übertragbares Agens in den untersuchten Geschwülsten anzunehmen.

Aus einer der Leukämien, die wir durch virushaltige Geschwulstfiltrate des Mäuseimpftumors Sa I erzeugt haben, konnten wir, ähnlich anderen Autoren[5,6], ein Virus *in vitro* isolieren und züchten[7]. Es lag deshalb nahe, die Wirkung beider virushaltiger Extrakte, der leukämogenen Geschwulstfiltrate und der Kulturflüssigkeiten des *in vitro* gehaltenen Virus, auf die Aktivität der Plasma-LDH zu untersuchen.

Methodik. Vom Tier wurden Viruslösungen durch Herstellung zellfreier Filtrate aus dem Mäuseascitestumor Sa I, wie früher angegeben, gewonnen[3]. Aus virushaltigen Suspensionskulturen von L-Zellen wurden auf dem Höhepunkt der cytopathogenen Veränderungen Zellhomogenate hergestellt und deren Überstand nach Zentrifugierung bei 1560 g für 20 min verwendet. Aus normalen virusfreien Kulturen von L-Zellen wurden ebensolche Homogenatüberstände für Kontrollversuche hergestellt. 3 Versuchsgruppen ausgewachsener Händlermäuse wurden mit jeweils 0,2–0,5 ml einer der Lösungen intraperitoneal beimpft. 48–72 h nach Beimpfung erfolgte die Blutentnahme teils durch Herzpunktion, teils durch Orbitalentnahme. Im abzentrifugierten Plasma wurde die LDH-Aktivität durch den DPN-H-Test im UV-Spektrophotometer (Eppendorf) gemessen und in Einheiten nach Bücher berechnet (Tabelle).

Es fand sich eine Erhöhung der LDH-Werte nach Gaben von Geschwulstfiltraten auf das 3fache der Norm im

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