was made to measure the pH very close to the plant. The plants were removed from their jars and given a quick shake, and the film of water on their surface was centrifuged off. The pH was found to be 0.5–1.0 above that of the surrounding medium and of the whole crushed plant. The pH of the epidermal cells must presumably be even higher, as the cell walls contain calcium pectate. Ruthenium red gave the characteristic stain even after heating sections for one hour in 2% ammonia at 90° C, which would remove any pectic acid or pectin.

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The Inhibitory Effects of Sorbose on Fungi[1]

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The fungi vary greatly in their ability to utilize different sugars as sources of carbon. Few fungi have been reported as being unable to utilize glucose or maltose (*1–3*), both of which are common, naturally occurring sugars. The utilization by certain fungi of other common sugars has been studied extensively (*4–9*). On the other hand, little is known of the utilization of the so-called rare sugars and of those that are not common in nature. Sorbose is one of the latter. It is formed from sorbitol, by the action of *Acetobacter suboxydans* (*10*). When present in a medium as the sole source of carbon, sorbose is utilized well by some fungi but only poorly or not at all by other species.

An unusual inhibitory effect of sorbose on the utilization of an available sugar, such as glucose, maltose, or sucrose, was discovered when certain fungi were grown in media containing a combination of sugars. The results of these studies are presented below.

The basal medium used was composed of asparagine 2 g, KH_2PO_4 1.0 g, $MgSO_4 \cdot 7H_2O$ 0.5 g, microelements as sulfates, Fe 0.2 mg, Mn 0.1 mg, Zn 0.2 mg, thiamine 100 µg, biotin 5 µg, and double-distilled water, 1,000 ml. Unless otherwise stated the amount of each sugar was 20 g/l. The media were adjusted to a pH of 6.0 (except in the study of the effects of pH), dispensed in 25-ml lots into 250-ml Erlenmeyer flasks, and autoclaved at 15 psi for 15 min.

The media were then inoculated, using uniform amounts of actively growing mycelium on agar, and thus eliminating any possible effects resulting from restricted or delayed spore germination. The cultures were incubated in a constant-temperature room at 25° C, except in temperature experiments, when refrigerator-incubators and water-jacketed incubators were used. Replicates of 4–10 cultures were used, and 2–6 cultures were harvested at the same time. Harvests of mycelium were accomplished by filtering the excess liquid through a fine cloth, drying the mycelial mats at 90° C for 12 hr, and weighing.

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TABLE 1

GROWTH OF 10 FUNGI IN 25-ML LIQUID MEDIA CONTAINING SINGLE SUGARS OR MIXTURES OF SUGARS
(Av dry wt of 2–4 cultures grown at 25° C)

Fungus	Days	Glucose	Glucose-sorbose	Maltose	Maltose-sorbose	Sorbose	Glucose (40 g/l)
Ceratostomella fimbriata	5	113	T*	84	T*	T*	107
Chaetomium globosum	12	373	292	364	25	T*	375
Alternaria solani	8	99	81	140	33	20	196
Sphaeropsis malorum	5	153	140	166	61	T*	240
Endothia parasitica	9	201	281	191	162	11	167
Choaneophora cucurbitarum	5	75	104	44	46	T*	100
Polyporus versicolor	9	152	232	126	192	47	221
Aspergillus rugulosus	13	239	439	200	221	189	448
Botrytis cinerea	9	182	266	210	310	198	379
Fusarium tracheiphilum	4	209	218	205	247	183	264

* T = trace of growth, estimated as less than 10 mg.

When a number of fungi were grown on sorbose, alone or in the presence of other sugars, it was noted that the response varied greatly. In general, the fungi fell into three groups: (1) growth on maltose greatly inhibited by the presence of sorbose, with little or no growth on sorbose alone; (2) growth on maltose-sorbose medium approximately the same as on maltose alone, but growth quite poor on sorbose alone; (3) fair to good growth on sorbose, as compared to maltose and maltose-sorbose media. The growth of representative fungi on different sugars is shown in Table 1.

C. fimbriata showed the greatest inhibition by sorbose. Under no condition did the cultures make more than 10–15 mg of mycelium in media containing 20 g sorbose/l, even in the presence of glucose or maltose. Additional experiments not reported here showed that smaller amounts of sorbose in the presence of glucose or maltose caused less inhibition. It is apparent that the presence of sorbose in the medium actually interferes with the absorption or the utilization of glucose and maltose by some fungi. It is also evident from Table 1 that the inhibition due to sorbose is greater in combination with maltose than with glucose.

The effect of temperature was then determined by culturing fungi previously shown to be inhibited by sorbose at temperatures ranging from 15° to 35° C.

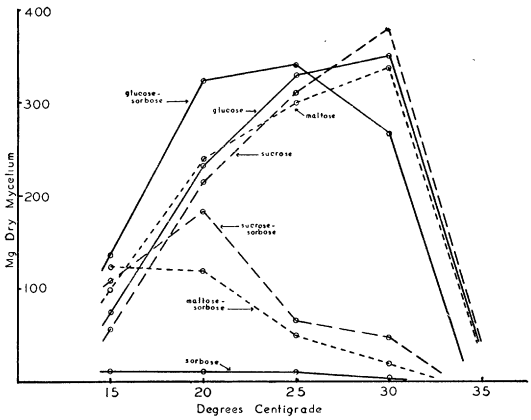

FIG. 1. The effects of temperature and the presence of sorbose on the growth of *C. globosum*.

The growth of *C. globosum* under these conditions is shown in Fig. 1. This figure shows that sorbose alone is not utilized, for no more than a trace of growth was produced at any temperature. There was little or no difference in the utilization of glucose, sucrose, or maltose when used alone. When these three curves are compared with the growth curves in media with these sugars plus sorbose, two general facts are revealed: (1) The inhibition of utilization of sucrose and maltose is greater than that of glucose. In fact, greater growth occurred on glucose-sorbose than on glucose at the lower temperatures. (2) The inhibition is increased with an increase in temperature from 20° to 25° and 30° C.

Other species, including *Sphaeropsis malorum, Sordaria fimicola,* and *Alternaria solani,* showed the same general response to increased temperature, but the degree of inhibition varied with the species. *C. fimbriata* failed to make more than a trace of growth in any medium containing sorbose at any temperature.

Fig. 1 also shows that the optimum temperature for growth of *C. globosum* is definitely dependent upon the sugars in the medium. In the absence of the inhibitory action of sorbose, growth was most rapid at 30° C. The increased inhibition of sorbose at this temperature more than balances, however, the tendency for increased growth. The net result in a mixture of sugars containing sorbose is more rapid growth at 20° C than at 30° C.

The effect of the hydrogen ion concentration of the medium can be considered only briefly. Media were prepared with the initial pH adjusted to 3.0, 4.0, 5.0, 6.0, and 7.0. *C. globosum* and *S. fimicola* grew on maltose media with the initial pH as low as 4.0. On maltose-sorbose media, growth of both fungi occurred at initial pH of 5.0 and above, but not at 4.0 or 3.0. Although the pH limits favoring growth in sorbose media appear to be narrower than those for growth in the absence of sorbose, these limits are not sufficiently narrow to account for the poor growth in the presence of sorbose.

One obvious effect of the presence of sorbose in the medium was the change in the type of growth. In sorbose media the colonies often remained separate, rounded, and even pelletlike, whereas in the absence of sorbose the mycelium was extensive. Microscopic examination revealed that mycelium growing in sorbose media was excessively branched. The most severe inhibition occurred in sorbose media, in which only traces of growth were present. Not only was hyphal extension inhibited, but many of the tips, particularly the apical cells, were killed. Staining the mycelium lightly with phloxine permitted counting the dead and living hyphal tips. *C. fimbriata* showed 77% and 15% dead hyphal tips in sorbose medium and maltose medium, respectively. In the same order and in the same media, 50% and 23% of the hyphal tips of *C. globosum* were dead. *A. solani* showed but few dead tips in sorbose medium, but excessive branching was evident.

It is thus evident that the presence of sorbose, a sugar poorly utilized by many fungi, may inhibit the utilization of a second sugar which alone is readily utilized. The reasons for this inhibition are not clear. The effect of temperature indicates that it might be based on enzyme activity or an absorption process. On the other hand, the killing of a high percentage of the hyphal tips suggests a toxic action of sorbose.

This study of sorbose utilization and inhibition of growth by sorbose is being extended, and further work is in progress. The results will be published elsewhere.

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Rapid Acclimatization of Insects to Anoxia, with Special Reference to the Housefly

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In studying the effects of rapid ("explosive") decompression on insects and certain related forms, the writers have observed that insects form a tolerance to anoxia when repeatedly decompressed. Acclimatization appears to be an anoxic response, as is shown by the fact that repeated exposures to a nitrogen atmosphere also effectively produce a tolerance. Moreover, it has been observed that a cross-tolerance can exist between nitrogen anoxia and decompression anoxia such that a preliminary exposure to either influence will create tolerance to the other. Thus, if a housefly (or possibly any insect) is exposed for 3 min to explosive decompression at 0.2–0.15 mm Hg pressure and 10 min later is exposed to an atmosphere