

the same time, maintains electrical continuity. The unit can be clamped in a ring stand or installed in a cross-member built over the cage. The arrangement is particularly useful in experiments involving extended periods of uninterrupted recording (for example, studies of drug effects).

The unit consists of a Plexiglas insert fitted into a ball bearing inside a fixed plastic shell. The wires in the recording cable are connected to the lower end of pieces of insulated stainless-steel tubing (hypodermic needle stock) passing through the insert. At the top these tubes extend upward from the axial hole in the shell. The upper end of each tube is bent to form an inverted "L," and its tip projects into an individual, mercury-filled, circular groove in the top of the shell. Each groove is separately connected with the amplifier of an electroencephalograph or some other recording device through a horizontal contact screw.

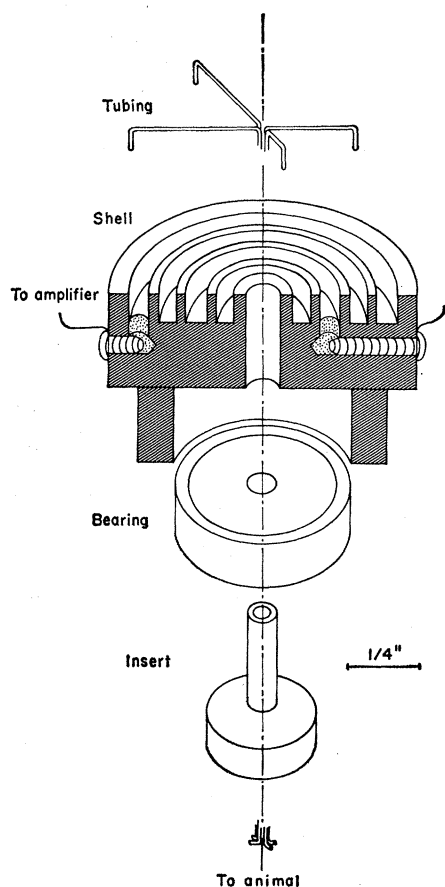


Fig. 1. Exploded view of cable coupler. Wires from recording cable are attached to bottom of tubes. A rubber band to support a loop of recording cable may be attached to the base of the insert. Leads to the recorder are connected to horizontal screws beneath grooves in the shell.

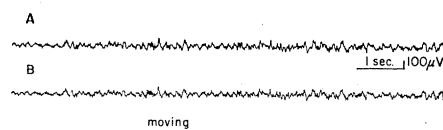


Fig. 2. Electroencephalogram from a bipolar electrode in rat prepyriform cortex. (A) Recording leads through cable coupler. (B) Leads directly from electrode to amplifier.

When the coupler is in use, torque on the recording cable is transmitted to the bearing. As it turns, the arms of steel tubing move through the mercury in the grooves, so that electrical continuity is maintained while the cable freely follows the animal's movement. Twisting or circling by the animal does not produce kinks in the cable. Figure 2 emphasizes the similarity of records made simultaneously through the coupler and bypassing it. There is no evidence of attenuation or distortion of the signal which passes through the coupler, nor are there distortions attributable to the coupler when movement occurs.

Care in construction will minimize both electrical and mechanical problems. Any of several plastic materials is suitable for the shell, so long as the dielectric quality is adequate. The diameter of the vertical orifice joining a groove in the shell and the hole for its contact screw should be the width of the groove; the mercury will not flow readily into a smaller hole because of high surface tension. The horizontal holes for the contact screws should accommodate large screws (6-32). The screws and the tubes from the recording cable should be stainless steel. Oxidation or amalgamation renders most other metals unsuitable.

Friction in the ball bearing is a potential problem, but this difficulty is minimized by using a precision bearing. The Plexiglas insert is press-fitted to the bearing, which in turn may be either press-fitted into the shell or held in place with screws. Since friction between the tubes and the walls of the grooves would hinder efficient operation of the coupler, the lengths of both limbs of each "L" must be measured accurately. Precise alignment is most easily obtained if the tubes are bent and cemented together before they are placed in the bearing insert. The resulting assemblage can then be accurately positioned for cementing in the insert.

It is helpful to employ a recording

cable which is long enough to form a small loop suspended by a rubber band attached to the bearing insert. This arrangement provides both leverage to turn the bearing and a means for removing slack in the cable when the animal moves about the cage.

Care in protecting the unit from dust prolongs its efficiency. Both to eliminate contamination of the mercury by dust and for physical protection of the stainless-steel arms, a slip-on cap can be fitted over the shell (1).

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Ascorbic Acid in the Nutrition of Plant-Feeding Insects

Abstract. *Bollworms, Heliothis zea (Boddie), and salt-marsh caterpillars, Estigmene acrea (Drury) gradually decreased in ascorbic acid content as they matured, even in its presence. Cotton leafworms, Alabama argillacea (Hübner), also lost ascorbic acid, although a dietary need for the vitamin was not proved. Pink bollworms, Pectinophora gossypiella (Saunders), reared without the vitamin, increased in ascorbic acid content as they matured, an indication that the vitamin was synthesized by the insect.*

Ascorbic acid is an indispensable nutrient for several insects (1, 2). All insects known to need a dietary source of the vitamin feed on plants. Common insects such as cockroaches, house flies, and mealworms have been reared in the laboratory for many years on simple diets without added ascorbic acid. The cockroach, *Periplaneta americana* (L.), can synthesize this vitamin (3). Analysis of a number of different species of insects showed ascorbic acid in the tissues of all (4). Although the exact requirements for this vitamin have not been determined, ascorbic acid is now being added to many diets used in rearing plant-feeding insects.

The boll weevil, *Anthonomus grandis* Boheman, requires ascorbic acid (2). When adult weevils were fed an ascorbic

acid free diet, the percentage hatch of eggs was greatly reduced, and when eggs hatched, the larvae did not survive. Larvae of the bollworm, *Heliothis zea* (Boddie), and the salt-marsh caterpillar, *Estigmene acrea* (Drury), failed to grow without dietary ascorbic acid.

Additional evidence that a dietary need for ascorbic acid exists in plant-feeding insects has been established. The ascorbic acid content of aseptically reared bollworms and several other insects was determined at different stages of their life cycles. The composition and preparation of the purified diets, the surface sterilization of the eggs, and the method of rearing the insects were essentially the same as reported previously (2). Soybean protein was substituted for half the casein in the diet. Ascorbic acid sterilized by filtration was added to the heat-sterilized diet at 40°C, just before it was dispensed into the rearing vials.

Ascorbic acid content of the diet was determined by titration with 2,6-dichlorophenolindophenol; insects and plant tissue were analyzed by the 2,4-dinitrophenylhydrazine method of Roe and Kuether (2). Sample weights of larvae, pupae, and moths ranged from 0.5 to 2.0 g and egg samples were about 100 mg. Most determinations were made in duplicate.

Table 1. Ascorbic acid in bollworms reared from purified diet.

Stage	Age (days)	Ascorbic acid content of bollworms ($\mu\text{g/g}$)	
		From diet*	B
Larvae	5 (0.12 g) [†]	532	
	6 (0.16 g) [†]		40
	7 (0.32 g) [†]	347	
	9 (0.63 g) [†]	163	
Prepupae	11-13	146	12
Pupae ♀	0	161	20
	1	125	
	3	145	
	5		0
Pupae ♂	6	162	
	7	110	
	8	122	
	0	112	13
	1	106	
	3	97	+
	4	85	
	5	114	0
Moths ♀ ‡	8	85	
	9	82	
		122	
		145	
Eggs §		421	
Eggs §		459	

* Diet A contained 0.4 g of ascorbic acid/100 g and diet B, 0.1 g/100 g, initially. † Average weight of larvae. ‡ Moths fed honey; eggs from these moths. § Moths fed ascorbic acid in honey; eggs from these moths.

Wild bollworms and those reared on a diet were analyzed at different stages of growth. Pink bollworms were reared on wheat-germ diet, as described by Adkisson *et al.* (5). Other insects were obtained from the field.

The mean ascorbic acid content of 5-, 7-, and 9-day-old larvae of the bollworm reared on a diet with 0.4 g of ascorbic acid per 100 g of food initially, was 532, 347, and 163 μg per gram of fresh insect tissue, respectively (Table 1). Little change in content occurred during the pupal period when no food was ingested, but a slight drop occurred just before emergence. When the insect was fed on a diet containing 0.1 g of ascorbic acid per 100 g initially, 6-day larvae contained only 40 μg of the vitamin per gram. The amount dropped to 20 μg when larvae became pupae. No ascorbic acid was detected in 5- to 6-day pupae. The percentage of larvae that became adults also was reduced when the initial concentrations of ascorbic acid were lower. Larvae reared on diets without ascorbic acid usually did not survive to the pupal stage. No ascorbic acid was found in these larvae.

Adults reared on a purified diet and fed honey contained 122 μg , and their eggs 145 μg of ascorbic acid per gram, as compared to 235 μg for field-collected moths. When moths were fed honey and ascorbic acid, the amount of the vitamin increased to 421 $\mu\text{g/g}$ of moths and to 459 $\mu\text{g/g}$ of their eggs, or four times the amount in moths fed honey only or in their eggs.

It should be pointed out that the bollworm does not receive a diet containing 0.4 g of ascorbic acid per 100 g under natural conditions. The large amount added to the diet was necessary to compensate for gradual loss during incubation at 29°C for 10 days. With an original content of 0.4 g, about 20 percent remained when the larvae had finished feeding, an amount comparable to that supplied in fresh plant material. At 0.1 g per 100 g of the diet nearly all the vitamin was lost by the 7th day, thus depriving the larvae of ascorbic acid during the last 3 days of their feeding period.

Ascorbic acid analyses of bollworms reared under natural conditions are shown in Table 2. Last-stage larvae from cotton fruit contained 340 μg and from sweet corn, 206 μg as compared with 163 $\mu\text{g/g}$ for those on a diet, as shown in Table 1. Adult moths from unknown sources, collected in a light trap, contained 192 μg and their eggs 235 μg of the vitamin per gram.

Table 2. Ascorbic acid content of several insects.

Stage	Food	Ascorbic acid ($\mu\text{g/g}$)
<i>Bollworm</i>		
Larvae (0.53 g) *	Cotton	340
Larvae (0.05 g) *	Corn	251
Larvae (0.15 g) *	Corn	228
Larvae (0.61 g) *	Corn	206
Moths ♀	Unknown	192
Moths ♂	Unknown	193
Eggs	Unknown	235
<i>Cotton leafworm</i>		
Larvae	Cotton leaves	254
Pupae (new)	Cotton leaves	153
Pupae (5 day)	Cotton leaves	125
Moths ♀	Cotton leaves	118
<i>Salt-marsh caterpillar</i>		
Pupae ♀	Cotton leaves	237
Pupae ♀	Diet	238
<i>Pink bollworm</i>		
Larvae	Diet	61
Pupae (new)	Diet	64
Pupae (4-6 day)	Diet	88
Pupae (6-9 day)	Diet	95
Eggs	Diet	181

* Average weight of larvae.

Cotton fruits contained about 0.05 g per 100 g and cotton nectar gave a positive test. Corn silks contained about 0.1 g per 100 g. Corn kernels were not analyzed.

Since normal yields of adults and eggs were obtained from diets containing 0.4 g of ascorbic acid, it was assumed that this quantity was sufficient for rearing under the experimental conditions. However, the ascorbic acid content of the eggs from field-collected moths (three samples) was higher than that from eggs of laboratory-reared moths (two samples), probably because the wild moths drank nectars that contained ascorbic acid. Thus, moths also should receive dietary ascorbic acid.

No substitute for ascorbic acid has yet been found. Cysteine and tocopherol do not alleviate the deficiency (2). On a diet free of ascorbic acid, few bollworms developed to the pupal stage and no moths were obtained. Pupae from diets with ascorbic acid weighed about 0.4 g and yields of adults were 70 percent or more. When araboascorbic acid was substituted for ascorbic acid, pupae weighed 0.26 g, the same as those obtained from a diet containing no ascorbic acid. The yield of adults was 39 percent, an indication that the isomer partially replaced the natural vitamin, especially in permitting growth. However, adults were abnormal, did not lay eggs, and survived only 1 to 3 days. Deficiency symptoms were characterized by darkening of body fluid and leakage of it at the

joints; the insect had difficulty molting and the wings and legs were brittle and broke off easily. These abnormalities have not been observed in studies of other nutrient deficiencies.

The ascorbic acid content of other plant-feeding insects is also shown in Table 2. The amount of the vitamin in cotton leafworms [*Alabama argillacea* (Hübner)] changed from 254 $\mu\text{g/g}$ of larvae to 118 $\mu\text{g/g}$ of moths. Adults probably are provided with ascorbic acid in their natural food. Salt-marsh pupae from cotton leaves and from a purified diet contained about 238 $\mu\text{g/g}$. Earlier studies showed that larvae did not grow without ascorbic acid (2). Larvae of the pink bollworm, *Pectinophora gossypiella* (Saunders), reared on a diet without ascorbic acid contained 61 μg , new pupae 64 μg , 4- to 6-day-old pupae 88 μg , 6- to 9-day-old pupae 95 μg , and eggs 181 μg of ascorbic acid per gram. This insect has been reared for many generations on such diets and apparently is capable of synthesizing all of this vitamin it needs. Apparently insects that require dietary ascorbic acid decrease in ascorbic acid content as they mature whereas the content of the vitamin in the pink bollworm increases.

All insects discussed here belong to the order Lepidoptera and must receive enough nutrients during the 8- to 14-day larval period to survive to the adult stage. Except for the pink bollworm, which prefers the seed, all these Lepidoptera eat the parts of plants rich in ascorbic acid. The bollworm eats the leaves and fruits of many plants. The salt-marsh caterpillar also feeds on many plants but eats only leaves, and the pink bollworm eats only cotton fruits. With the exception of the salt-marsh caterpillar, adults of these insects feed on plant juices that undoubtedly contain ascorbic acid.

Our present data and that contained in an earlier report (2) clearly demonstrate a nutritional role for ascorbic acid. To what degree this vitamin influences feeding is not yet known. Ito (6) reported that ascorbic acid acts both as a phago-stimulant and a nutrient for the silkworm.

Our present knowledge indicates that the number of plant-feeding insects requiring an exogenous source of ascorbic acid exceeds the number able to synthesize it. Since this vitamin is apparently of vital importance to reproduction, studies including more than one generation of an insect may be necessary to determine the requirement.

Discovery of the dietary role of ascorbic acid makes it possible to rear plant-feeding insects in the laboratory on defined diets. Furthermore, the metabolic function of ascorbic acid can be studied in a system in which the concentration of the vitamin can be controlled (7).

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Coesite and Stishovite: Stepwise Reversal Transformations

Abstract. *Very marked differences in the metastable persistence of coesite and stishovite have been demonstrated; the former possibly persists indefinitely below 1000°C at 1 atmosphere "dry," and the latter completely decomposes in minutes above 500° to 600°C to an amorphous or short range order phase. Quartz was grown (metastably) at temperatures well above its stability field from both coesite and stishovite, possibly by way of a short range order phase. The absence of stishovite in meteor-impact craters cannot be taken as evidence that it was not formed. If it has "reversed" in normal natural environments the product would almost certainly be a short range order phase or derivative.*

In all the reconstructive transformations which the silica minerals undergo, it has long been assumed that an amorphous or short range order (SRO) intermediate phase is formed as the first step. Recent data (1) substantiate this idea. Specifically, short range order is the most general term that can be used to describe phases which are not crystalline (that is, they do not have periodicity over distances greater than about 100 Å). The extent of structural order in SRO phases is therefore limited to the first sphere of coordination and possibly up to a few unit cells.

The SRO phase of a one-component system may have properties dependent on the conditions of treatment, while in a multi-component system the composition of the SRO phase also may vary.

In spite of the existence of the SRO as an intermediate in such transformations it is clear, however, that structural control derived from the parent or original modification is still a reality. While Ostwald's step rule (2) may have constituted an overgeneralization, its fundamental validity is rooted in the transmission of inherent structure in a direct manner through epitaxy or topotaxy, or in a more indirect manner through an SRO intermediate.

With the increasing use of coesite and stishovite (3, 4), which are the high pressure modifications of SiO_2 as indicators of meteorite impact, it is essential to know the conditions under

which these forms would revert to other forms of SiO_2 , and the structural controls and kinetics of the various reactions that occur.

Quenching experiments and high-temperature x-ray measurements were used to follow the reversal of the high-pressure phases, coesite and stishovite, to the various forms of SiO_2 which are stable at 1 atmosphere.

The quartz-coesite equilibrium (5) focused our attention on the kinetics of this reaction. In an effort to determine the activation volume for the coesite-quartz reaction, an attempt was made to determine how long the metastable coesite would persist at atmospheric pressure in air at various temperatures. Such data would also give information on the possible ceramic use of coesite. Coes (6), in his original paper, stated

Table 1. Results of heating coesite. The pressure was one atmosphere.

Temp (deg)	Time (hr)	Ratio† cristo- balite/ coesite	R.I.		
			Aggre- gate	Min.	Max.
1165	20	0:1	1.555	1.483	1.585
1155	24	2:1	1.485	1.476	1.560
1160	42	1:0	1.476	1.470	1.496
1340	1	0:1‡	1.553	1.500	1.573
1345	2	0:1‡	1.517	1.480	1.557
1340	5	1:1‡	1.480	1.466	1.553
1070	550	0:1	1.585	1.580	1.587

* ± 0.002 . † The ratio of cristobalite to coesite is based on x-ray diffraction intensities. ‡ Both the microscope and x-ray diffraction show traces of quartz. For comparison, the normal refractive indices are cristobalite (1.487, 1.483); quartz, (1.544, 1.553); coesite (1.598 average). Data for 17 μ coesite.