acid and riboflavin, used alone or in combination, and the effect of low-temperature treatment, prior to application of the vitamin or vitamins, on early growth and accumulation of dry matter in plants. It is concluded that ascorbic acid or riboflavin, or both, at low concentrations are definitely more growth-promoting for the vernalized than for the unvernalized seeds (*5*).

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Interactions of Pectin and Protein in the Heat Coagulation of Proteins

Abstract. Pectin decreases the heat coagulation of many proteins, including the soluble proteins of pea stems, ovalbumin, and bovine serum albumin. This observation probably explains the decrease in heat coagulation of proteins from pea stems following in vivo auxin administration, since auxin causes a great increase in the soluble pectin content of treated cells.

We have reported previously (*1*) that the administration of 2,4-dichlorophenoxyacetic acid or other auxins to growing pea stem tissue results in a decreased heat coagulability of the soluble proteins of the particle-free homogenate of the stem. We now believe this effect to be due to an increase induced by auxin administration in the level of cold water soluble pectins (*2*) which then interact with the proteins during heat coagulation.

Subapical sections from the stems of green or etiolated peas were harvested, homogenized in 0.001*M* EDTA (ethylenediamine tetraacetate) plus 0.5*M* sucrose at *p*H 6.0, and centrifuged as previously described (*1*). The clear particle-free supernatant fraction was used for the heat coagulation experiments and also for assays of the pectin which is soluble in cold water. Pectin was determined by a slight modification of the method of McComb and McCready (*3*), in which pectin is precipitated by the action of 70 or 95 percent ethanol for 72 hours at 2°C, demethylated in 0.05*N* NaOH for 30 minutes, and hydrolyzed by heating for 10 minutes with concentrated H_2SO_4 to the free galacturonic acid, which was then determined colorimetrically with carbazole. Because of the lack of specificity, this method was checked several times against a hydrolytic procedure involving purified polygalacturonase (*4*). Furthermore, the in vivo synthesis of the material assayed by the above procedures was inhibited by $10^{-3}M$ ethionine, and this inhibition was reversed by methionine. Both facts give strong support to the view that the material assayed was pectin.

Table 1 shows the increase in the cold water soluble pectin content of green pea stem sections which had been incubated for 20 hours in the light in $10^{-5}M$ 2,4-dichlorophenoxyacetic acid (2,4-D), 1 percent sucrose, and 0.01*M* phosphate buffer, *p*H 6.1. It is clear that the growth increase induced by 2,4-D is accompanied by a massive synthesis of pectins, as well as by the decreased heat coagulability of proteins previously reported (*1*).

We next investigated the possibility that the deliberate addition of pectin in vitro could accomplish the same effects on protein coagulation as did the in vivo administration of 2,4-D. To the clear particle-free homogenate of pea stem were added varying quantities of commercial citrus pectin. Aliquots (5 ml) in test tubes were immersed in a boiling water bath for 10 minutes. The heat coagulum was centrifuged at 3000*g*, washed twice with cold distilled water, poured into a previously tared aluminum weighing dish and dried overnight at 90°C. Table 2 shows the relation between pectin concentration and the dry weight of the precipitate so obtained. This effect of pectins in reducing the heat coagulability of pea proteins could be observed only over the rather narrow range *p*H 5.5 to 6.5, as shown in Table 3. The inhibitory effect of the pectins disappears below *p*H 5.0, where ionization of pectin carboxyls is repressed, and above *p*H 7.0, where pea proteins fail to coagulate by heat.

Similar experiments were undertaken with various commercially available proteins, including bovine serum albumin, and with freshly prepared ovalbumin. Typical results are shown in Fig. 1. While these curves differ in detail, they both show that pectins inhibit heat coagulation in the general range *p*H 5.3 to 5.7, and promote heat

Table 1. Effect of the auxin, 2,4–dichlorophenoxyacetic acid, on growth and the content of cold water soluble pectin in sections of green pea stems.

2,4–D (mole/liter)	Increase in fresh wt. (%)	Uronic acid equiv. (μg/g)	
		In original fresh wt.	In final fresh wt.
0	46	349	239
10^{-5}	124	870	382

Table 2. Effect of commercial citrus pectin on heat coagulation of pea stem proteins at *p*H 6.0

Pectin concn. (μg/ml)	Coagulum (mg dry wt.)
0	4.9
6.6	4.6
13.3	4.5
20.0	3.4
26.6	1.4
100.0	0.2

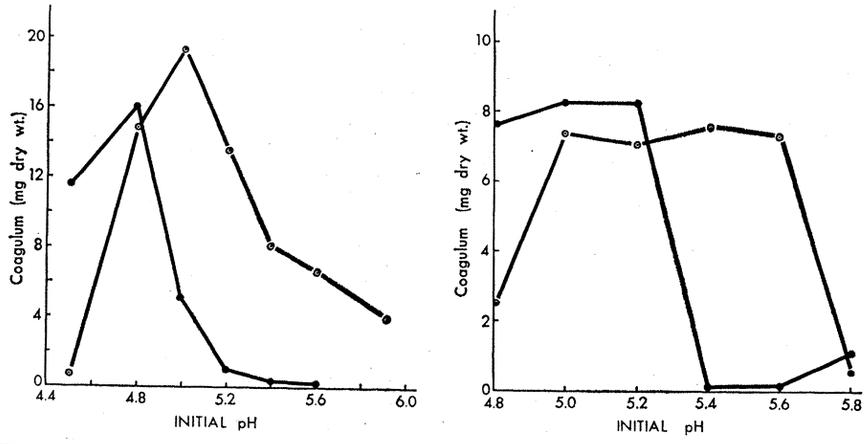

Fig. 1. Effect of *p*H on the heat coagulability of bovine serum albumin (left) and ovalbumin (right) in the absence (open circles) and presence (solid circles) of commercial citrus pectin (100 μg ml).

Table 3. Effect of *p*H on inhibition by pectin of heat coagulation of protein from pea stem.

*p*H	Coagulum (mg dry wt.)	
	Without pectin	With 100 μg/ml pectin
5.0	2.8	2.3
5.5	2.7	0.0
6.0	2.5	0.2
6.5	2.1	0.3
7.0	0.2	0.2

coagulation in the lower *p*H range. If the citrus pectin was boiled before it was added to the homogenate, there was no effect on protein coagulation.

From the quantitative data on ovalbumin and bovine serum albumin, whose molecular weights are known, minimal effective molar ratios were calculated for the interaction of pectin and protein. These calculations indicate that considerably less than one pectin molecule is required for interaction with one protein molecule. However, in view of the polydisperse nature of the commercial pectin employed, additional purification is required before an exact ratio can be calculated.

Although pectins are generally believed to be localized in cell walls, they may in fact also be present in the cytoplasm. If they are, the interaction of pectin and protein described here might be physiologically significant and could explain the effects of auxin on cytoplasmic viscosity and on cyclosis (*5*). It must be noted also that the fraction here designated as "cold water soluble pectin," after Albersheim and Bonner (*2*), has not been rigorously demonstrated to be polymerized methyl galacturonate, but could as well be a hemicellulose (*6*).

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Embryogenesis of the Human Temporomandibular Joint

Abstract. The structures of this articulation were found to originate from two different blastemata situated at some distance from each other and operating at different rates and opposing directions. A condylar blastema evolving dorsally contributes to the formation of the condylar cartilage, the disc, the aponeurosis of the external pterygoid muscle, and the capsular elements of the lower joint level, while a temporal blastema develops the articular structures of the upper level in a forward direction. At the end of the fourth fetal month all joint elements including a glenoid fossa and articular eminence are present in their primitive form.

Recently experimental evidence has been presented to show that the condylar cartilage growth center, in contrast to epiphyseal plates of long bones, responded to induced stresses by adaptive growth movements (*1*). This unique behavior of condylar cartilage was thought to be related to its ontogenetic pattern whose details, however, are little known (*2*). In order to assess it systematically in adequate human material, the heads of 25 human fetuses of ascending stages from 15 mm CR (crown to rump) length to full term have been prepared for histologic analysis in either the coronal or sagittal plane (6 μ paraffin serial sections).

The condylar blastema appears in the form of a mesenchymal cell condensation at the dorsal end of the dentary in the 24 mm fetus. This bilateral anlage of the future mandible, the dentary, starts to ossify in the symphyseal integument at the 19 mm stage. In the 35 mm fetus it has developed dorsally and medially, in relationship to the Meckel's cartilage and the primordium of the external pterygoid muscle (Fig. 1). At the 55 mm stage, it produces an osseous head (Fig. 2) and matures at the 60 mm stage when condylar cartilage is differentiated. Simultaneously secondary cartilage also appears at the sites of the aponeuroses of the masticatory muscles. This growth spurt of the mandible coincides chronologically with that of the prechordal segment of the cranial base (*3*). While a paramedian portion of the Meckel's cartilage is incorporated into the mandibular body by a process of endochondral ossification and the secondary cartilages also disappear, the condylar cartilage starts endochondral ossification at the 85 mm stage to become henceforth the growth center of the mandible.

The evolution of the temporal blastema lags in time and space behind the condylar blastema. The first anlage is seen at the 35 mm stage in connection with the intramembranous ossification of a squamotemporal bone rod (Fig 1). At the 50 mm stage, in the region of the auditory meatus, the first temporal joint structures are differentiated in the

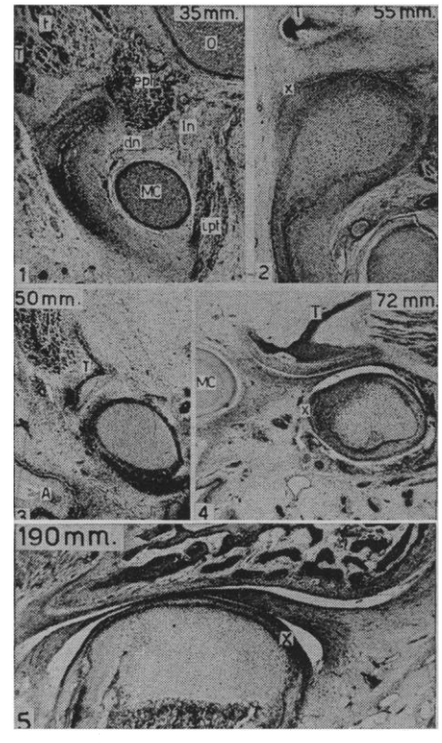

Fig. 1. Coronal section of a 35 mm CR human fetus shows condylar blastema at the level of the orbitosphenoid cartilage (*O*) and above the Meckel's cartilage (*MC*). Note beginning of intramembranous ossification of squamotemporal rod (*T*); *t*, temporal muscle; *ept*, external pterygoid muscle; *ipt*, internal pterygoid muscle; *ln*, lingual nerve; *dn*, inferior dental nerve (× 56). Fig. 2. Coronal section of a 55 mm CR human fetus shows formation of an osseous condylar head. Incipient chondrogenesis is seen at the dorsolateral aspect (*x*). Note relative aplasia of temporal blastema (*T*) at this level (× 56). Fig. 3. Coronal section of a 50 mm CR human fetus shows evolution of the temporal blastema (*T*) situated dorsally to the condylar blastema (*T*) at the level of the external auditory meatus (*A*) (× 56). Fig. 4. A sagittal section of a 72 mm human fetus shows that the temporal joint elements develop in a forward direction and the condylar elements, including the lower glenoid cavity and disc, in a backward direction as indicated by (*x*), center of most active chondrogenesis (× 36). Fig. 5. In a sagittal section of a 190 mm human fetus all articular elements are present. Condylar cartilage proliferation proceeds dorsally (*x*) while the temporal joint structures develop in a forward direction (× 36).