value of the loss tangent varies somewhat from sample to sample but the range we have observed is restricted to values between 0.001 and 0.02. It is somewhat difficult to understand the nature of this frequency-independent loss tangent but it is a well-recognized property of many technical dielectrics and is common in dry materials (*8*). It appears that exceedingly dry, powdered rock samples at frequencies of 10^3 hz or higher are likely to have a loss tangent that is nearly frequency-independent.

We can readily convert the data on loss tangent into an equivalent attenuation distance by noting that skin depth is given by $\sqrt{\sigma\mu\omega/2}$, where σ is the conductivity, μ is the permeability, and ω is the rotational frequency. The conductivity can be replaced by $\varepsilon''\omega$, where ε'' is the imaginary part of the dielectric constant. The loss tangent, tan δ, is given as $\varepsilon''/\varepsilon'$, where ε' is the real part of the dielectric constant. Curves of the attenuation distance as a function of frequency for values of K tan δ, where K is the relative dielectric constant, are given in Fig. 2. The measured samples have values of K tan δ ranging from 0.003 to 0.06 at frequencies up to 1 Mhz.

Direct observations of the lunar surface have been tabulated by Weaver (*9*) for the frequency range from 10 to 75 Ghz. When the attenuation depths are plotted as a function of frequency, they give a close fit to the curve for K tan $\delta = 0.003$. This value is similar to that observed for samples at lower frequencies. Tyler (*10*) used reflections from the communications channel of the Explorer 35 satellite to make an estimate of the loss tangent at 136 Mhz. This gives essentially the same value for K tan δ. The available information on absorption in the lunar surface layers over a frequency range from 136 Mhz to 75 Ghz seems to be consistent with the existence of dry, dielectric materials. There may be some resonance absorption phenomena present but the available data give no indication of this.

If this observation is correct, it implies that the uppermost lunar layer has a loss tangent of about 0.001, a value close to that of dry, powdered natural samples in the laboratory. Many samples of dry, powdered rocks exhibit data like those shown in Fig. 1. The d-c electrical conductivity of these samples at 27°C ranges from 10^{-13} to 10^{-16} mho/m. It seems probable, therefore, that the d-c conductivity of the uppermost layers is exceedingly low. Typical penetration depths that might be expected at other frequencies can be estimated from Fig. 2, although both K and tan δ tend to increase at lower frequencies.

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Hormonal Stimulation of Lactose Synthetase in Mammary Carcinoma

Abstract. *A transplantable rat mammary carcinoma (R3230AC) synthesizes significant quantities of the mammary gland enzyme lactose synthetase in the immature virgin female rat. In this hormonal environment, mammary glands do not synthesize the enzyme. Prolactin further stimulates the enzyme activity in the tumors to levels found only in mammary glands of rats in late pregnancy or during lactation.*

The enzyme that catalyzes the final rate-limiting step in the biosynthesis of lactose, lactose synthetase (UDP-galactose : D-glucose-1-galactosyltransferase; E.C. 2.4.1.22), has two protein components (*1*). The A protein is a galactosyltransferase which catalyzes the synthesis of *N*-acetyllactosamine from UDP- (uridine diphosphate) galactose and *N*-acetylglucosamine (*2*). The B protein is alpha lactalbumin which behaves as a "specifier protein" and modifies the substrate specificity of the A protein from *N*-acetylglucosamine to glucose, so that lactose synthesis results (*3*). In normal mammary tissue, synthesis of both A and B protein can be stimulated by prolactin (*4*), but during pregnancy placental progesterone inhibits the synthesis of the B protein and thus prevents the synthesis of lactose until parturition (*5*).

Since this complex regulatory mechanism is integrated with the overall differentiation process in mammary tissue, it is pertinent to determine whether mammary carcinoma tissue is sufficiently differentiated to synthesize lactose synthetase and utilize this hormonal regulatory system. We now show that appreciable A and B protein activities are present in rat mammary carcinoma tissue and, furthermore, that the levels of A and B activity are stimulated by prolactin.

A spontaneous transplantable rat mammary adenocarcinoma (R3230AC) has been carried in vivo in this laboratory for 18 months in intact virgin female Fisher rats. Perphenazine (5 mg/kg), a potent stimulus for pituitary prolactin release (*6*), or ovine prolactin itself (1 mg per day) was given to groups of tumor-bearing virgin female Fisher rats. They were killed at various times after hormone treatment, and the inguinal mammary glands and tumors were removed for assay of A and B protein activities by a radioactivity method (see *2*).

Table 1 presents the A and B protein activities in immature rat mammary glands, before and after four consecutive days of perphenazine administration, and in adult lactating glands. The low enzyme activities in the unstimulated

Table 1. The A and B protein activities of lactose synthetase in mammary tissue of immature rats, immature rats treated with perphenazine, and lactating rats. The results are expressed in nanomoles per minute per milligram of protein (mean ± standard error of the mean). Five animals were used for each experiment.

UDP-galactose utilized	
A activity	B activity
Immature	
0.043 ± 0.015	0.014 ± 0.004
Immature, 4 days after perphenazine	
0.054 ± 0.014	0.086 ± 0.040
Lactating	
0.97 ± 0.26	2.38 ± 0.15

glands and the high activities in the lactating glands are consistent with previous observations (*4*). The negligible response of enzyme activity to perphenazine administration is surprising in that during this 4-day period there is unequivocal histologic evidence of differentiation and increased protein and lipid synthesis in the mammary glands (*6*).

In contrast to the results noted above, the level of A protein activity in the tumors was high relative to that in the mammary glands of untreated rats (Table 2). The enzyme level was further increased after administration of estradiol, prolactin, or perphenazine. It is not known whether the estradiol acted directly on the tumor tissue or indirectly by stimulating pituitary prolactin release.

The B protein activity in the tumors from untreated rats was also very high relative to the activity in the untreated mammary glands (Table 2). Perphenazine or prolactin further stimulated B protein activity to levels found only in adult lactating mammary glands. Adrenocorticotropin, bovine growth hormone, or progesterone did not influence the level of A or B protein activity in the tumors.

It seems probable that in these experiments the virgin mammary tissue was not sufficiently differentiated to respond to the 4-day prolactin stimulus by synthesizing lactose. A period of intense hormonal stimulation, such as pregnancy, is required for complete differentiation of these cells. Hence, the experiments on the mammary carcinoma cells are of particular interest because, even in immature unstimulated rats, the tumors synthesize appreciable quantities of this unique mammary gland enzyme, lactose synthetase. The relatively high activities may be the result of normal endogenous prolactin secretion since they are further increased by exogenous prolactin or by the perphenazine-induced release of endogenous prolactin.

This tumor cell can be thought of as being in a functional state of differentiation comparable to the normal mammary cell in late pregnancy or early lactation with respect to A and B protein synthesis. Enzymes earlier in the biosynthesis pathway of lactose (hexokinase, uridine diphosphate galactose-4-epimerase, and uridine diphosphate glucose pyrophosphorylase) have been measured in carcinogen-induced mammary tumors (*7*). The activities of these enzymes are also comparable to those in normal glands in late pregnancy or early lactation.

Although the biochemical abnormalities responsible for sustained growth in cancer cells have not been identified, the R3230AC cell demonstrates certain features of hormone-dependent control of growth. If estrogen is administered to the tumor-bearing rat or if a prolactin-secreting pituitary is transplanted into these animals, the rate of tumor growth is markedly reduced in spite of increased protein and lipid synthesis. Also, if normal endogenous amounts of pituitary hormones are eliminated by hypophysectomy, tumor growth as well as specific protein and lipid synthesis is inhibited (*8*).

The ability of the R3230AC tumor cell to retain these certain, very specialized, differentiated characteristics (A and B protein synthesis and regulation) in addition to hormone-dependent growth regulation provides an experimental model of the mechanism of hormonal regulation of growth and differentiation of mammary carcinoma cells.

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Table 2. Effect of hormone treatments on A and B protein activity in R3230AC mammary carcinoma. The rats received the indicated treatments subcutaneously on days 1 and 2 and were killed on day 3. The results (nanomoles per minute per milligram of protein) are expressed as the mean of five separate tumors ± standard error of the mean.

Treatment	Dose	UDP-galactose utilized	
		A protein	B protein
Saline alone		0.127 ± 0.017	0.675 ± 0.022
Adrenocorticotropin	5 units	0.159 ± 0.009	0.562 ± 0.045
Bovine growth hormone	1 mg	0.131 ± 0.022	0.542 ± 0.032
Estradiol	20 μg	0.200 ± 0.015	0.662 ± 0.020
Progesterone	4 mg	0.128 ± 0.016	0.470 ± 0.075
Ovine prolactin	1 mg	0.213 ± 0.018	1.440 ± 0.209
Perphenazine	0.5 mg	0.336 ± 0.047	1.150 ± 0.130

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Lymphocyte Stimulation: Transfer of Cellular Hypersensitivity to Antigen in vitro

Abstract. *Tuberculin-sensitive human lymphocytes cultured for 36 hours with tuberculin elaborate a soluble material which causes nonsensitive lymphocytes to respond to tuberculin in vitro by transformation and proliferation.*

When lymphocytes from an individual with delayed hypersensitivity to an antigen are placed in short-term tissue culture with that antigen, they transform into blastlike cells, synthesize DNA, and divide (*1*). A small percentage of peripheral blood lymphocytes initially responds to such antigenic stimulation and, after approximately 48 hours, undergoes a proliferative response for several generations (*2*). Cells arising from this proliferation account for the majority of the lymphoblasts present when cultures are examined on days 5 to 7 (*2*). In a population containing sensitive cells, the recruitment of lymphocytes not themselves initially sensitive to antigen might serve, particularly during the first 2 days of culture, as an additional mechanism to enlarge this hypersensitive response of lymphocytes. There is evidence that this response to antigen in vitro parallels and has several characteristics of delayed hypersensitivity in vivo (*3*).