in these experiments were known to be over a year old. Clearly the learning of new flight calls is not limited to a critical period in the first year of life. Absence of an age-limited critical period permits a bird, throughout its lifetime, to learn new calls as its social environment changes.

Call and song learning may also differ in the nature of their reinforcement. Flight-call learning may occur in the context of courtship feeding, a behavior frequently seen during integration of flocks and pairs. Within a bout of courtship feeding, the two participating individuals frequently utter their characteristic flight calls. If courtship feeding is associated with the learning of new calls, then social factors or possibly even food (*6*) may serve as a source of the external reinforcement. Although social or emotional factors have been suggested as possible sources of reinforcement in song learning in some highly social passerines (*8*), there remain many examples of song learning that occur in the absence of any obvious external reinforcement (*3*).

In addition to providing evidence for individual recognition, learning, and the absence of a critical period, these data demonstrate the ability of finches to imitate. Taken as a whole the data provide support for the hypothesis (*9*) that avian vocal imitation is sometimes employed for recognizing and maintaining contact with other members of the species, and particularly with the mate (*10*). Related to this, Thorpe believes the ability of parrots and mynahs to mimic man may be biologically understandable if in nature these birds employ their mimetic powers for recognizing species members and maintaining contact with them (*11*). We do not as yet know if parrots and mynahs do in fact imitate conspecifics as part of their social behavior, but since the data reported here indicate that goldfinches and siskins do it seems pertinent to point out that the canary, a close relative of goldfinches and siskins, is also known to produce crude renditions of human speech (*12*). Thus, like the pine siskin's mimicry of a European siskin flight call, avian mimicry of the human voice may be a manifestation of interspecific social integration occurring in unnatural circumstances.

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5. The American goldfinch and pine siskin are often placed in the genus *Spinus*. Regardless of problems of nomenclature, ornithologists agree that the pine and European siskins are more closely related to each other than either is to the American goldfinch.
6. What is called "courtship feeding" in carduline finches varies from a brief touching of bill tips to prolonged bouts involving the insertion of bills and the transfer of food. I have only observed the transfer of food during the nesting season. Courtship feeding is usually considered as a component of reproductive behavior; however, it is observed throughout the year in flocks of captive carduelines and it can involve a pair of males or females as well as a heterosexual pair. This suggests that courtship feeding has social functions beyond those of reproduction.
7. In ten tests the control flight call was presented first; in four tests the mate's flight call was played first. The control male was a neighboring male in some tests, a strange male in other tests. All playbacks were done in the field with the speakers and the observer located about 10 m from the nest.
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Metobromuron: Acetylation of the Aniline Moiety as a Detoxification Mechanism

Abstract. p-*Bromoaniline is rapidly acetylated by four soil microorganisms. Two fungal species convert metobromuron to* p-*bromoacetanilide, but a bacterial and an algal species do not metabolize metobromuron. Acetylation may serve as a detoxification mechanism by competing with azobenzene formation in utilizing the aniline formed by metabolism of substituted urea herbicides.*

Chloroacylanilide herbicides are degraded in soil to the corresponding anilines, which in turn are oxidatively coupled to azobenzenes (*1*). We have observed an alternative pathway. We studied the bromine-containing herbicide 3-(*p*-bromophenyl)-1-methoxy-1-methylurea (metobromuron) at 10 times the concentrations recommended for field application, rather than the much higher concentrations (up to 1000 times the recommended rates) previously used. In contrast to the results with chlorine-substituted acylanilides, we found that the aniline moiety of metobromuron is not converted to an azobenzene by soil microorganisms but is instead acetylated to the less toxic acetanilide.

Pure culture studies were made of the soil microorganisms *Talaromyces wortmanii, Fusarium oxysporum,* and *Chlorella vulgaris* and of a *Bacillus* species. The organisms were incubated with 10 μg of [C_6H_6-^{14}C]metobromuron per milliliter of synthetic medium for 18 days at 24°C and aerated on a gyratory shaker. The parent compound and its metabolites were extracted with two volumes of chloroform. Partial purification was accomplished by filtering the chloroform extracts through a layered column of unactivated Florisil and charcoal. The resulting solutions were concentrated and analyzed by thin-layer chromatography [Brinkmann precoated TLC plates, silica gel F-254, developed with ethyl acetate; chromatograms developed with a mixture of chloroform and acetic acid (60 : 1, by volume)]. The location of radioactive spots on chromatograms was determined by autoradiography. The quantity of each radioactive compound was determined by scraping the spot into a scintillation solution and counting in a scintillation counter. The metabolites were identified on the basis of their chromatographic properties and their mass spectra. The latter were obtained by extracting TLC spots with ethyl acetate, evaporating the extracts in quartz capillaries, and introducing the capillaries into the solid inlet system of a Perkin-Elmer model 270 GC-DF mass spectrometer.

The extracts from the fungal cultures gave one TLC spot with an R_F value of 0.19, which was shown by its mass spectrum to be *p*-bromoacetanilide [molecular ion at *m/e* (mass to charge) 213/215; base peak, *m/e* 171/173 (C_6H_6BrN)+]. *Talaromyces wortmanii* converted more than 5 percent of the herbicide to *p*-bromoacetanilide, while a trace of the derivative was formed by *Fusarium oxysporum*. Neither of the other two organisms formed this derivative. In no case was *p*-bromoaniline or *p*-bromoazobenzene observed. To discount the possibility that acetylation of *p*-bromoaniline is an artifact of the TLC solvent system, a pure standard

of *p*-bromoaniline was spotted and run through the solvent system several times. There was no acetylation.

When the organisms were incubated with *p*-bromoaniline at concentrations of 1, 10, and 20 μg/ml, all of the organisms tested converted the aniline to the acetanilide. No starting aniline or the azobenzene was observed after incubation for 7 days. Thus, the acetylation of substituted anilines in soils may be competitive with the oxidative coupling to azobenzenes, at least in the case of bromine-containing aniline from the phenyl ureas. Unless the coupling reaction is very fast or the aniline concentration is very high, little or no azobenzene may be formed. The removal of the anilines from the soil by acetylation should be advantageous in the metabolic degradation of urea herbicides because the acetanilides are usually less toxic than either the corresponding aniline or azobenzene.

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Adenosine 3′,5′-Monophosphate, Adrenocorticotropic Hormone, and Adrenocortical Cytosol Protein Synthesis

Abstract. *In cultures of mouse adrenocortical tumor cells (Sato's minimal deviation Y-1 clonal strain), the acceleration of steroid biosynthesis after exposure to adrenocorticotropic hormone or cyclic adenosine 3′,5′-monophosphate is maximum within 15 to 60 minutes and precedes any significant increase in labeling of protein with [4,5-^{3}H]leucine. However, when cytosol proteins are separated by acrylamide-gel electrophoresis, rapid changes in the amount and labeling of several protein fractions are evident in less than 30 minutes and are no longer evident within 60 minutes. This finding supports the proposal that the effects of tropic hormones and their intracellular mediators involve rapid selective effects on protein synthesis.*

Adenosine 3′,5′-monophosphate (cyclic AMP) is the probable intracellular mediator of many of the actions of hormonal polypeptides and amines on their target cells. The role of the nucleotide in the steroidogenic effect of adrenocorticotropic hormone (ACTH) on adrenal cortex seems well established (*1*). The activity of membrane-associated adenyl cyclase is stimulated by ACTH, increasing the conversion of adenosine triphosphate to cyclic AMP. An increased concentration of cyclic AMP stimulates the enzymatic conversion of cholesterol to Δ^5-pregnenolone, the rate-limiting enzymatic step in adrenal steroidogenesis (*2*). The nature of the step or steps between the increase in cyclic AMP and the cleavage of the cholesterol side chain is still obscure.

That protein synthesis may be involved either directly or indirectly is suggested by the fact that (i) pharmacologic inhibitors of protein synthesis can interfere with the rapid effect of ACTH or cyclic AMP on steroid biosynthesis (*3*) without preventing the action of ACTH on adenyl cyclase (*4*), and (ii) there is an apparent lack of a direct effect of cyclic AMP on a side-chain cleaving system in mitochondria (*5*). This evidence has been difficult to evaluate because there has been no proof that the action of the inhibitors was exclusively derived from decreased protein synthesis (*6*). In addition, there has been no previous evidence that ACTH affects adrenocortical protein synthesis during the period of its initial effects on steroidogenesis. Evidence that ACTH and cyclic AMP actions involve protein synthesis must first include such a demonstration; second, cyclic AMP must be shown to have an ACTH-like effect on side-chain cleavage in adrenocortical mitochondria.

We studied the effects of exogenous ACTH and cyclic AMP on a clonal strain of mouse adrenocortical tumor cells (*7*) in monolayer culture. These cells respond to ACTH with a rapid increase in adenyl cyclase activity (*8*) and to both ACTH and cyclic AMP with a rapid increase in steroidogenesis (*9*). We added 5 munit of ACTH or 1.0 μmole of cyclic AMP per milliliter of tissue culture medium, amounts which produced roughly the same steroidogenic effect in cultures incubated for 2 hours or less. The effect of ACTH on the rate of steroid production [number of micrograms of the major steroidal products (*9*) per 10^6 cells per hour] was maximum in less than 30 minutes (Fig. 1). The difference in time of the peak effect of cyclic AMP from that of ACTH was accompanied by extreme variability of the steroidogenic rate in experiments where the cultures were incubated with cyclic AMP, and can be attributed to the relative impermeability of most plasma cell membranes to the nucleotide (*1*).

To investigate alterations in protein synthesis during the acceleration of steroid synthesis after addition of ACTH or cyclic AMP, we also added 2.5 μc of [^{3}H]leucine per milliliter of tissue culture medium (*10*). Tumor cells were harvested in an ice-cold saline solution, rinsed three times to wash away medium proteins, subjected to osmotic shock, and homogenized; they

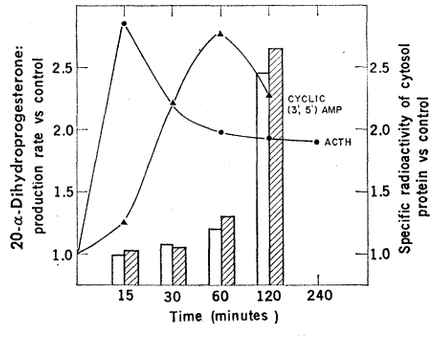

Fig. 1. Effects of ACTH and cyclic AMP on steroid production and on the specific [^{3}H]leucine radioactivity of cytosol protein in adrenocortical tumor cells. Closed circles indicate the ratios of 20-dihydroprogesterone production (micrograms of steroid per 10^6 cells per unit time) in cells exposed to 5 munit of ACTH per milliliter of culture for various periods (*9*). Closed triangles indicate those exposed to 1.0 μmole of cyclic AMP per milliliter. Bars indicate the ratios of the specific radioactivity of the cytosol protein in cells treated with ACTH to untreated cells (open bars), and of cells treated with cyclic AMP to controls (closed bars). The values shown represent averages from five different experiments.