

with their proper designations as well as HeLa contaminants which masquerade under the same name.

It is, no doubt, true that the different bona fide strains of HeLa perform in different ways and exhibit many distinct characteristics; the same is true of cultures of HeLa that are known by different designations, but are de facto HeLa strains themselves. Nevertheless, these strains have retained the characteristics that we have tabulated here, in spite of different passage levels under different growth conditions and in different laboratories. It is this group which we specifically addressed here and the one which has led many investigators to believe that "HeLa by many other names can spell trouble."

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Phytoestrogens: Adverse Effects on Reproduction in California Quail

Abstract. *Phytoestrogens, largely formononetin and genistein, are produced in the leaves of stunted desert annuals in a dry year. When ingested by California quail, these compounds apparently inhibit reproduction and prevent the production of young that will not have adequate food. In a wet year, forbs grow vigorously and phytoestrogenic substances are largely absent. Quail then breed prolifically and the abundant seed crop carries the enlarged population through the winter.*

The California quail, *Lophortyx californicus*, breeds irregularly in the more arid portions of its range, depending on the amount of winter rainfall preceding the spring nesting season. When rainfall is generous, a rich carpet of forbs which supplies both greens and seeds for quail consumption is produced. Under this circumstance the birds breed vigorously. In a relatively dry year, the ground may be sparsely covered with stunted forbs and annual grasses; then quail breeding is desultory, and few or no young are produced. There seems to be some direct connection between forb growth and the breeding suc-

cess of quail (1-3), and it has long been presumed that the control is nutritional. The breeding success of arid-land quail has been linked to storage in their livers of vitamin A obtained from green foods (4, 5). Whereas vitamin A is necessary for quail, there is no clear relation between the availability of this food supplement and the reproductive vigor in the birds (6). We surmise that there are other nutritional components of green food that regulate reproduction.

Phytoestrogens in subterranean clover inhibit breeding in domestic sheep (7). Thus, this group of compounds might reg-

Table 1. Effect of diet on egg production in three pairs of California quails.

Diet	Feeding period (month/day)	Onset of egg laying	Eggs per pair (No.)
Turkey starter*	10 March to 6 June	8 April	62
Low energy, low protein†	3 March to 30 June	8 May	28
Turkey starter plus subclover extract‡	3 March to 30 June	2 June	12

*Turkey starter contained 26 percent crude protein. †Low protein diet contained 15 percent crude protein. ‡Subclover extract contained biochanin A, genistein, and formononetin.

ulate quail breeding. We analyzed leaf samples of various plants eaten by quail and identified four isoflavones—biochanin A, genistein, daidzein, and formononetin—plus coumestrol, in several of them. The extraction procedure was that of Beck (8), and identification of the compounds was made by thin-layer chromatography by means of the Eastman chromatogram with silica gel incorporated with a fluorescent indicator. The relative amount of each phytoestrogen present in a sample can be estimated (low, medium, high) by the degree of fluorescence under ultraviolet light. Absolute amounts cannot be measured by this method.

A feeding trial was initiated to test the effect of phytoestrogens on egg production. Penned quail fed turkey starter mash plus subclover extract (10 ml of extract with 500 g of food) laid eggs 2 months after the control pair started laying, and fewer eggs were produced (Table 1). The subclover extract contained high amounts of biochanin A and genistein and a medium amount of formononetin.

Wild quail were collected near Shandon, San Luis Obispo County, California, to ascertain the occurrence of phytoestrogens in the crop preceding and during the breeding season. Beginning in December 1971, birds were shot each month throughout the 1972 and 1973 breeding seasons. Crop samples for a given month were pooled; part of the material was analyzed for identification of foods consumed and part was homogenized for analysis of phytoestrogen content. The year 1972 was dry with scant forb growth and poor breeding success among the quail (25 immature offspring per 100 adults in the fall), whereas 1973 was wet with good forb growth and excellent quail breeding (325 immature offspring per 100 adults).

The most apparent difference between 1972 and 1973 is the continuing appearance of formononetin and genistein in the foods consumed in 1972 and the near absence of these compounds in the spring and summer foods taken in 1973 (Table 2). Daidzein was not detected in either year. There appears to be an inverse correlation between the incidence of estrogenic substances in the diet and the success of reproduction in the quail.

In the dry 1972 season there was an adequate supply of forb and grass leaves available for consumption by the quail, but the plants were small and stunted and the birds ingested them in only modest amounts (Table 3). By contrast, the green growth in 1973 was robust, and quail consumed large quantities of greens and insects as well. The quail paired and nested both years; but in 1972 reproduction lagged, and many of the birds gave up trying to produce a brood

Table 2. Phytoestrogen content of pooled quail crops (both sexes) taken during a winter and spring (1971 to 1972) that led to poor reproduction (25 immature offspring per 100 adults) and the following year (1972 to 1973) that led to high reproduction (325 immature offspring per 100 adults). Coumestrol was found at low concentration in the samples for 3 and 4 June 1972 and September 1973 and at high concentration in the sample for June 1973. Concentrations are listed as low (L), medium (M), or high (H).

Collection date	Crops (No.)	Phytoestrogen concentrations		
		Biochanin A	Formononetin	Genistein
1971				
12 December	4	L		
13 December	5			M
28 to 31 December	5		L	
1972				
17 January	4	L	L	L
22 to 24 January	12	L	L	L
28 January	18		M	L
4 to 5 March	5		M	M
27 to 30 March	4		L	L
16 April	3		L	L
13 to 14 May	3	L	L	L
13 to 14 May	6		L	
3 to 4 June	3		L	L
July	6			
August	4			
12 December	7			M
1973				
3 to 5 January	9			H
5 to 27 January	10			L
30 March	7			
April	2		L	
June	5			
July	6			
August	3			
September	6			

Table 3. Green leafage content of 86 quail crops taken during the 1972 and 1973 breeding seasons. The balance of the diet (last two lines) consisted of insects and seeds of about 60 species of forbs and grains, not here itemized.

Substance	Amount (percent by volume) by season					
	Winter* 1971-72	Spring 1972	Summer 1972	Winter 1972-73	Spring 1973	Summer 1973
Filaree (<i>Erodium</i>)	6.2	1.8	tr†	31.0	8.2	tr
Forbs (unidentified)	12.2	8.2	8.0	14.8	8.1	0.2
Grass blades (Graminae)	8.3	tr		19.5	3.9	
Clover (<i>Trifolium</i>)	tr	0.8	0.7	tr	17.5	
Miner's lettuce (<i>Montia</i>)					7.1	
Lotus (<i>Lotus</i>)				3.8	1.1	
Lupine (<i>Lupinus</i>)				3.5	0.3	
Wild carrot (Umbelliferae)					3.2	
Insects	tr	5.7	tr	tr	7.6	10.2
Seeds	73.3	83.5	91.3	27.4	43.0	89.6

*Winter means December to February; subsequent seasons are also 3 months each.

†Trace amount.

and gathered into winter coveys as early as June. In 1973, there was persistent nesting and renesting through the summer, and young were still being hatched as late as September. We presume that stunted forbs contain more isoflavones than the same plants growing vigorously and even when taken in modest amounts these compounds seem to inhibit reproduction.

The luxuriant forb growth of 1973 produced abundant seeds which served to nourish the young through the subsequent year. In 1972, the seed crop probably would have been inadequate to support a large crop of young, had they been produced. The presence or absence of phy-

toestrogens in the foliage of annual plants may be one of the mechanisms that coordinates quail reproduction with the prospective available food resource.

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Drought in the Sahara: Insufficient Biogeophysical Feedback?

In a recent report (1) Charney *et al.* used a global general circulation model to test the hypothesis that overgrazing in the Sahelian zone might be, at least in part, responsible for the recent drought in that region. They claimed that their results supported this hypothesis.

A major criticism of their work is that, although they have taken account of the vegetation's effect on albedo, they have completely ignored its effect on evapotranspiration. Even though vegetated surfaces often absorb more radiation than bare ground, they are usually cooler because much of the absorbed energy is used to evaporate water. It is also regrettable that Charney *et al.* continue to subscribe to the idea that localized land and vegetation management can have a significant effect on rainfall climate.

In view of the scarcity of published data on Saharan albedo, the value of 0.35 used by Charney *et al.* does not appear to be unrealistic (2). The albedo postulated for a vegetated Sahara (0.14) also appears to be reasonable (3). However, this could vary considerably with type of vegetation, ground cover, and moisture status. Although it is true that a decrease in the albedo of a surface will cause an increase in the net radiation income, for a decrease effected by adding live vegetation one must expect that at least part of the extra energy will be used in transpiration. The addition of water to the atmosphere, through transpiration, probably would not enhance precipitation since atmospheric moisture is already quite high (4) and the effect of a slight augmentation is not likely to be great (5).

If the albedo change is effected through the use of vegetation, as implied in Charney *et al.*, transpiration will be a necessary consideration and a change in the

surface energy budget can be expected. Van Bavel and Fritschen (6) compared energy balances over dry and wet soils and found that, although the daily net radiation was about 20 percent greater over the wet soil, the sensible heat flux to the atmosphere and the surface temperature were both greater for the dry soil. I carried out a comparison of two adjacent areas in a natural savanna region of northeastern Uganda (7); one area was heavily grazed, and the other was protected from grazing (Table 1). These data show the grazed surface, in spite of its higher albedo and lower net radiation, to be considerably warmer during the daytime than the ungrazed surface. Thus, protection from overgrazing might be expected to reduce convection and precipitation rather than to increase them.

If the Saharan albedo change could be effected by artificial means, so as to avoid transpiration, then the results of Charney *et al.* would likely apply. This case also deserves comment. Figure 2 of Charney *et al.* shows an increase in rainfall to the north of 16°N for the lower albedo case but a decrease south of this latitude. Examination of a population distribution map (8) shows that there is a very low population density between 16°N and 25°N compared with the very high density between the equator and 16°N. In addition, an article in the *World Meteorological Organization Bulletin* (4) describes the Sudano-Sahelian zone as lying "approximately between latitudes 10°N and 20°N." Thus, even without transpiration, the effect of decreased albedo appears to be a reduction of rainfall in the most heavily populated region of the Sahel.

The presentation of the results in Charney *et al.* also invites criticism. The choice of the month of July and the use of

latitudinal averages obscure pertinent spatial and temporal variations. Although the precipitation pattern depends most strongly on latitude, there is some variation with longitude, particularly south of 10°N (8). Whereas the summer months are the wettest in the latitudes 10°N to 15°N, farther north autumn and winter are the rainiest seasons (9). Therefore, one must not assume that the effect on annual precipitation would be the same as that simulated for the July precipitation, particularly in the more northerly part of the zone.

As a final point, the July rainfall values indicated by Charney *et al.* (in their figure 2) for the region north of 15°N appear to be far higher than those reported elsewhere (10).

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Ripley's criticisms are apposite. We are grateful for the opportunity they provide to clarify the physics of our model and to discuss further the implications and limitations of our results. His major criticism is that, while considering the effect of changes in vegetative cover on albedo, we have completely ignored the effect of vegetation on evapotranspiration. He points out that vegetated surfaces are usually cooler than bare ground because much of the absorbed solar energy is used to evaporate water, and he concludes from this that protection from overgrazing might be expected to lower surface temperatures and thereby reduce rather than increase convection and precipitation. We agree with his premises but not with his conclusions. It is true that the very primitive hydrology

Table 1. Mean daily energy balance data for 4 days (0700 to 1900, local time) for a savanna area in northeastern Uganda in January 1964 (5). The effective surface temperature values were obtained from radiation measurements, assuming an emissivity of 1.0.

Area	Global radiation (joule cm ⁻²)	Net radiation (joule cm ⁻²)	Albedo	Effective surface temperature (°C)
Grazed savanna	2340	1256	0.20	34.7°
Ungrazed savanna	2340	1549	0.15	29.2°