ent in 2 samples of lyophilized nucleoprotein fraction. The DNA is probably an inactive contaminant in the procedure, since culture tests on both low and high polymer DNA from various sources showed no stimulating effect.

Summary. 1. A quantity isolation method is described for a chick embryo nucleoprotein fraction based on von Euler and Heller's(10) procedure for liver nucleoproteins. 2. When this fraction was tested in an assay medium of undialyzd chicken plasma and dilute undialyzed horse serum large outgrowths of chick heart fibroblasts were obtained with well-defined alterations in cell morphology and mode of outgrowth. 3. The nucleoprotein fraction described contains protein, 10.3% PNA, 0.3% DNA and has a characteristic nucleoprotein absorption spectrum. 4. High polymer pentose nucleic acid isolated according to Nishioka and Ibuka(9) produced no changes in size or appearance of assay cultures.

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L-Lyxoflavin in the Starting Diets of Turkeys and Chickens.* (20369)

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Since Pallares and Garza(1) described the isolation of L-lyxoflavin there has been considerable interest in this compound in nutrition although there is still comparatively little published information concerning its essentiality in the diet. Emerson and Folkers(2),

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working with hyperthyroid rats, reported that L-lyxoflavin had a growth promoting activity distinct from that obtained with various liver fractions. Wahlstrom and Johnson(3,4) used L-lyxoflavin in a synthetic milk for pigs and reported highly significant growth increases at 7 weeks. Bruins *et al.*(5) obtained a growth increase of about 10% when L-lyxoflavin was fed to chicks for an experimental period of 2 weeks. Their diet was of a purified type and contained riboflavin. Cooperman *et al.* (6), using the ration described by Bruins *et al.*(5), obtained a small response from feeding supplemental L-lyxoflavin, but no more than was obtained by the addition of supplemental riboflavin at equivalent levels. Shorb (7) reported that L-lyxoflavin inhibited riboflavin competitively in the growth of *L. casei* in a riboflavin assay medium, but at high levels of riboflavin the L-lyxoflavin stimulated the growth of the organism. Ershoff(8) has reported that L-lyxoflavin is ineffective as an antithyrotoxic factor for rats although various liver substances were effective.

Because of the conflicting results with this compound reported in the literature, we are presenting our studies. No previous results with turkeys have been reported. A number of experiments have been conducted in this laboratory using L-lyxoflavin in experimental diets for both turkeys and chickens.

Experimental methods. All experiments were started with day-old poults or chicks and conducted for a period of approximately 4 weeks. Each experimental group consisted of 10 birds raised in standard electrically-heated batteries, with feed and water being supplied *ad libitum*. The turkey poults used were Broad Breasted Bronze of medium fast-growing strains. The ration used in the turkey work was a corn-soybean oil meal type ration described by Sherwood and Briggs(9) modified by the addition of 0.1% choline chloride, 10 μg vit. B_{12} and 10 mg procaine penicillin G per kg of ration. This ration calculates to be adequate in all other B-complex vitamins. In view of the recent report by Cooperman *et al.*(6) data from a separate experiment are shown in Table I which indicate that the basal ration was not improved by the inclusion of additional riboflavin or riboflavin rich materials. The purified diet used in the chick trials was as follows: 59.2% glucose, 25% Alpha protein,§ 4% clarified corn oil, 6% salts 3M,‖ 5% cellulose, 0.6% DL-methionine, 0.2% choline chloride and the following vitamins (in mg/kg): thiamine hydrochloride, 20; riboflavin, 8; calcium pantothenate, 20; pyridoxine hydrochloride, 6; biotin, 0.2; nicotinic acid, 100; folic acid, 2; vit. B_{12}, 0.01; alpha tocopherol acetate, 10; menadione, 1. Vit. A was added at the rate of 20,000 IU/kg; vit. D_3 750 ICU/kg; and procaine penicillin G, 15 mg/kg. The chicks used were New Hampshire birds of a medium fast-growing strain. Mortality in all groups was negligible. The L-lyxoflavin used was crystalline L-lyxoflavin obtained from Dr. Karl Folkers, Merck & Co., Inc., Rahway, N. J. We also used in one trial a concentrate containing 33 mg L-lyxoflavin/g.

TABLE I. Effect of Addition of Other Vitamins on 4 Week Weights of Turkeys.

Exp. No.	Supplement added	Sex	No. birds/ mortality	28 day wt
1	0	—	10/0	575
	Dried brewers yeast, 5%	—	20/0	564
2	0	♂	6/0	560
	Niacin, 40 mg/kg	♂	6/0	567
	Riboflavin, 25 mg/kg	♂	6/0	555

Results and discussion. The results of the experiments with turkeys are shown in Table II. In the first 2 experiments, in which only male birds were used, the differences in growth favored the poults fed the L-lyxoflavin-supplemented diet by a highly significant margin. The percentage increase resulting from the use of L-lyxoflavin was 12.0% and 17.6% in Exp. 1 and 2, respectively. In the third trial, in which poults of mixed sexes were used, the difference was 7.1%, but this difference was not significant at the 5% level. It is believed

§ Alpha protein, an isolated soybean protein manufactured by The Glidden Co., Chicago, Ill.

‖ Salts 2M (Hill, E. G., and Briggs, G. M., PROC. SOC. EXP. BIOL. AND MED., 1951, v76, 417) modified by replacing the ferric ammonium citrate with ferrous ammonium sulfate to give equivalent amounts of iron.

TABLE II. Growth Response of Poults Resulting from Adding L-Lyxoflavin to a Corn-Soybean Ration.

Exp. No.	L-lyxoflavin added, mg/kg	Sex	No. birds/ mortality	27 day wt (g)	Stand. error	F. E.*
1	0	♂	10/1	546	± 16.6	2.04
	25	♂	"	612†	± 17.4	1.79
2	0	♂	10/2	535	± 34.9	1.94
	35	♂	10/0	629†	± 20.8	1.84
3	0	♀ + ♂	"	537		1.92
	0	♀ + ♂	"	532	± 14.9§	1.95
	33‡	♀ + ♂	10/1	583		1.98
	33‡	♀ + ♂	11/1	572	± 16.5§	2.10

* F. E. = feed efficiency, or $\frac{\text{g feed consumed}}{\text{net g gained}}$.

† Wt increase compared to control, significant at 1% level by "t" test.
‡ Lyxoflavin added in form of concentrate.
§ Duplicate groups considered together.

TABLE III. Growth Response of Chicks Fed a Purified Diet Supplemented with L-Lyxoflavin. 13 experiments.

Series No.*	43		45		46		48	
Sex	♂ ♂		♀ + ♂		♀ + ♂		♂ ♂	
Supplement to basal	4 wk wt, g	S.E.†	4 wk wt, g	S.E.†	4 wk wt, g	S.E.†	4 wk wt, g	S.E.†
Basal alone	295	± 7.2	335	± 12.1	317	± 9.5	329	± 11.0
25 mg L-lyxoflavin/kg	306	± 6.6	331	± 7.2				
10% soybean oil meal	335	± 10.7	338	± 10.0				
Basal minus cellulose + 10% soybean oil meal			350	± 13.1				
As gp. 4 + 25 mg L-lyxoflavin/kg			360	± 10.5				
10% dried whey product + 25 mg L-lyxoflavin/kg					330	± 12.9		
10% torula yeast + 25 mg L-lyxoflavin/kg					324	± 10.6		
10% whey product + 10% torula yeast					335	± 7.4		
As gp. 8 + 25 mg L-lyxoflavin/kg					341	± 10.6		
5% torula yeast							333	± 9.7
As gp. 10 + 25 mg L-lyxoflavin/kg							342	± 8.4
As gp. 10 + 10 mg % orotic acid							349	± 6.8
As gp. 12 + 25 mg L-lyxoflavin/kg							336	± 8.3

* 20 New Hampshire chicks per group. † Stand. error of the mean.

that the smaller response in the third experiment was due to the use of birds of mixed sexes, as females do not usually grow as fast as males.

Feed efficiency was slightly improved when L-lyxoflavin was added to the diets of male poults, but this was not the case when poults of mixed sexes were used.

The results of the trials with male chicks are shown in Table III. In no case did the addition of L-lyxoflavin result in any growth improvement that could be considered significant. In this respect we have failed to corroborate the findings of Bruins *et al.*(5) even though the diet used in these experiments was similar. It will be noted that orotic acid also failed to give any growth response with this diet. Further, the combination of orotic acid, L-lyxoflavin and dried yeast failed to result in any growth response over the dried yeast alone or the dried yeast plus the L-lyxoflavin. Similarly in trials not presented in Table III no growth response was noted with 20 chicks fed a corn-soybean diet supplemented with L-lyxoflavin, crystalline riboflavin, vit. B_{12} and procaine penicillin G in 2 trials.

No explanation can be offered as yet for the small growth stimulating action of L-lyxoflavin. Emerson and Fokers(2) suggested that it was of vitamin nature; but before this

compound can be accepted as a vitamin, evidence for its existence in natural foods must be found. Before the growth-promoting effect of L-lyxoflavin can be considered of practical importance, studies must be made with this compound using diets of a more varied nature including a wider range of natural feed ingredients. Studies of this nature are at present in progress.

Summary. In trials with turkey poults highly significant growth responses were obtained with male poults fed a corn-soybean diet supplemented with L-lyxoflavin. Chicks fed purified diets failed to show a growth response when the diet was supplemented with L-lyxoflavin.

We wish to thank Merck and Co., Inc. for the L-lyxoflavin and crystalline vitamins, Lederle Laboratories for crystalline folic acid, and Professor T. H. Canfield for sexing the birds.

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Distribution of P^{32} Following the Injection of TEPA* in Rats.† (20370)

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The ethylenimines have been shown to be effective in producing regressions of certain transplantable tumors in rats(1-3). These compounds will inhibit growth and cause cellular destruction of the tumor tissue at concentrations which will not produce any demonstrable changes in normal tissue. At higher concentrations, however, they will inhibit the growth of the testes and white blood cells(2,3). Still higher concentrations will cause general tissue damage. Studies have been started, therefore, to determine the specific effects of these compounds upon the various tissues together with their distribution throughout the body. A preliminary report is presented here of the distribution in rats of P^{32} from N, N′, N″-triethylenephosphoramide (TEPA*).

Materials and methods. Fifty-eight Sprague-Dawley male rats (175-200 g) were divided into 3 groups, 2 of which were implanted subcutaneously with the Flexner-Jobling carcinoma, the third group serving as a control. Ten days after transplantation, each of 24 rats in Group A bearing the tumors received one dose subcutaneously of TEPA*, 5.0 mg/rat, (900 μg P/rat), (1.55×10^4 CPM/μg P). In Group B the 16 control rats were treated with TEPA* at the same dosage level as the rats in Group A. The 18 rats in Group C bearing the tumors were injected subcutaneously with Na_2HPO_4*, 4.3 mg/rat, (400 μg P/rat), (7.16×10^3 CPM/μg P). At 1, 2, 15, and 30 hours following injection of the tagged compound, several animals from each group were autopsied. Samples of liver,

* Radioactive.

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