Maine Agricultural Experiment Station
Department of Animal and Veterinary Sciences

Hatch Project No. _____

Effective July 1, 1972

I Title: Physiology of Low Level Intake of Polychlorinated Biphenyl Isomers by Chickens

II Justification: Polychlorinated biphenyls (PCBs) are widely distributed in the global environment. They have been prepared and used commercially since 1929. Although their production, sale and use has been drastically reduced in the last few years, the estimated production in 1971 was still about 20 thousand tons (Chem. & Eng. News 1971). Studies in the last few years have shown a wide environmental distribution for PCBs (Risebrough, et al., 1968). Carpenter and Smith (1972) found widespread concentrations of small pieces of brittle plastic in the western Sargasso Sea. These particles have surfaces which may serve for the attachment of diatoms and hydroids. They suggested that these plastics may be the source of the PCBs recently found in the tissues of oceanic animals. These facts coupled with the exceedingly long half-life of the PCBs suggest that small amounts of these materials may be found in the future in various kinds of foods and feeds.

Work to date with chickens has been done only with commercial PCBs marketed as Aroclors by Monsanto Chemical Co., specifically Aroclor 1242 and 1248, fed in relatively high concentrations for short periods of time. It would seem appropriate to study the effects of feeding minute amounts of Aroclor or specific PCB isomers to chickens over long periods of time.

III Previous work: The toxic effects of relatively large amounts of PCBs (Aroclor) fed to chickens has been reported, (McCune, et al., 1962, Rehfeld, et al., 1971, and Scott, et al., 1971) and in other species of birds (Risebrough, et al., 1968, and Delong, and Linder, 1971).

Rehfeld, et al., fed Aroclor 1248 to one-day old chicks for 4½ to 5 weeks. General edema, enlarged heart sacs and depression of secondary sexual characteristics were observed in chicks fed levels greater than 20 p.p.m.. Where 50 p.p.m. were fed in the diet, growth was depressed, mortality was 50% and some birds showed impaction of the crop and proventriculus indicating a temporary stasis of the digestive tract.

Scott, et al., (1971) fed PCBs (Aroclor 1248) to laying hens for a period of 8 weeks. PCBs were transferred to eggs, muscle tissue and body fat of the laying hens but had no effect on shell strength. Ten p.p.m in the diet reduced hatchability by 20% after 4 weeks and 44% after 8 weeks. Muscle tissue lost PCBs rapidly on withdrawal of the material from the diet. The level in body fat and in eggs remained at a high concentration up to 4 weeks after withdrawal of PCBs from the diet.

There is some indication that the toxicity of Aroclor is correlated with the chlorine content of the material (Rehfeld, et al., 1971). Thus, toxicity may be correlated with relative chlorine saturation of the biphenyl molecule which might indicate that the lower chlorine isomers are metabolized or turned over faster than are those containing more chlorine. Desaiah, et al., (1972) have shown that four PCBs in fish exhibit prominent inhibitory effects on oligomycin-insensitive Mg^{2+} ATPase, with that in muscle homeogenate being the most sensitive. Aroclors 1242 and 1254 were more inhibitory than 1221 and 1268.

According to a recent science editorial (Chem. Eng. News 1972) the PCB problem is far from resolved. It is reported that some European-made PCBs contain tetrachlorodibenzofurans which are highly toxic compounds and are suspected carcinogens. A nonpolychlorinated biphenyl has been found in the urine of rats fed 100 to 500 p.p.m. Aroclor 1254 and this biphenyl compound was found in the Aroclor administered. It was found to have a mass spectrum number (m/e) of 304. A contaminant with the same m/e number has been identified as tetrachloroparadibenzofuran in the French and West German products. However, the component m/e 304 could be tetrachloromethyl biphenyl since methyl biphenyls are present in trace concentrations in the

feedstock used to make PCBs. Rats fed the above Aroclor have enlarged livers and abnormalities in the liver cells. Whether the m/e 304 component contributed to the toxic effect is not known. The same type of liver enlargement has been observed in chicks (Rehfeld, et al., 1971).

PCBs appear to block the chick's response to Vitamin D. This may partially explain the involvement of PCBs in egg shell formation, contributing to thin shells.

Methodology using gas-liquid chromatography (GLC) for the determination of PCB still leaves much to be desired. "The pattern of GLC peaks from a sample containing PCB is often not exactly like that from any of the Aroclors. This is probably due to a combination of circumstances, e.g., weathering and/or metabolism of the residue; and perhaps variation in the recovery of the different PCB components through the methodology. Choosing an appropriate Aroclor reference against which to measure a residue requires good judgment on the part of the analyst." (Pesticide Anal. Manual Vol. I, Section 251.17). Thus, work needs to be done to standardize the methods of analysis for PCBs. Also, standards of reference must be employed which are consistent among laboratories.

References

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Risebrough, R.W., P. Rieche, D.B. Peakill, S.G. Herman and M.N. Kirven. 1968. Polychlorinated biphenyls in the global ecosystem. Nature 220: 1098-1102.

Scott, M.L., D.V. Vadehra and V.A. Mullenhoff, G.L. Rumsey and R.W. Rice. 1971. Results of experiments on the effects of PCBs on laying hen performance. Proc. Cornell Nutr. Conf. p.p. 56-64.

IV Objectives

1. To test the long term effects in chickens of continuously feeding diets containing small amounts of an Aroclor or pure polychlorinated biphenyl isomers.

2. To study assay procedures for the quantitative estimation of polychlorinated biphenyl isomers in feed ingredient, feeds and animal tissues.

3. To investigate methods for blocking the physiological activity of PCBs.

V Procedure

Male and female chickens of a meat type production strain will be used in both meat and egg production studies. They will be fed commercial type rations to which will be added either an Aroclor, a pure polychlorinated biphenyl isomer, or a combination of isomers at preliminary levels of 0, 0.01, 0.10, 1.00, and 10.0 p.p.m. Nanogram and picogram levels may be investigated depending on results with the preliminary levels.

The PCB materials which will be investigated are as follows: Aroclors 1232, 1242, 1248, 1254 and 1260. In addition, the initial isomers to be studied are:

2 - Chlorobiphenyl
4,4' dichlorobiphenyl
2,4 - dichlorobiphenyl
2,4,6 - Trichlorobiphenyl
2',3,4 - Trichlorobiphenyl
2,2',4,4' - Tetrachlorobiphenyl
2,3,4,5,6 - Pentachlorobiphenyl
2,2',4,4',6,6' - Hexachlorobiphenyl
2,2',3,3',5,5',6,6' - Octachlorobiphenyl
2,2',3,3',4,4',5,5',6,6' - Decachlorobiphenyl

Feed consumption, growth data, egg production and mortality records will be maintained. Tissue accumulation of PCBs will be followed by tissue analysis either following

slaughter of representative animals or by tissue biopsy techniques. Tissues which will be analyzed are liver, muscle and fat. PCB content of feces will be determined.

Any animal dying will be autopsied and appropriate gross and histological observations will be made.

Analysis for PCBs in tissues and eggs will conform to present official procedures outlined in Pesticide Analytical Manual, Vols. I and II, Food and Drug Administration, U.S. Department of Health, Education and Welfare, 1971, and the "compilation of PCB analytical procedures used by FDA laboratories," 1972, or their revisions.

Broiler studies will be for 8-week periods. Females used in egg production studies will be fed the test rations from day of hatch through the first laying year. Tests for fertility and hatchability of eggs from hens will be carried out. Also, the carry over of PCBs from egg to the baby chick will be investigated.

All animals, eggs and litter to be disposed of will be burned in an incinerator equipped with a stack burner to completely combust escaping gases.

The ability of the gut to transport PCBs will be investigated by the Crane and Wilson technique (1958) in which an everted section of gut is formed into a sac and incubated in a glucose buffer containing the material to be transported. This technique will permit the study of materials which might block the uptake of PCB by the intestine.

The data will be analyzed by appropriate statistical methods.

VI Cooperating Agencies

None.

VII Termination by June 30, 1977. Five (5) years.

VIII Financial Support

Salaries

Research Technician

Supplies

Polychlorinated biphenyl isomers

glassware

chemicals

Capital Equipment

Detector replacement

Gas Chromatograph

Laboratory Alterations

water lines

vacuum lines

exhaust fan

hood canopy

IX Personnel:

Francis H. Bird and Richard W. Gerry

X Institutional Unit:

Department of Animal and Veterinary Sciences

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