# PCBs in Laying Hens

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In the past year there have been several instances of contamination of poultry feedstuffs by PCBs. Accidental feeding of contaminated feed to commercial chicken breeders has resulted in decreased hatchability. There have been only a few controlled studies on the effects of PCBs on reproduction in chickens.

SLIDE 1 - In the experiments I am going to discuss today polychlorinated biphenyls were added to a standard breeder ration and fed ad libitum to mature White Leghorn chickens. The PCB diet was fed for 9 weeks. At the end of 9 weeks, the PCB diet was withdrawn and the birds were fed the breeder ration with no PCBs added.

Throughout the entire experiment feed consumption, body weight gain, mortality, egg weight, eggshell thickness, egg production, fertility and hatchability were followed. Unhatched embryos were examined for teratogenic effects. Survival and body weight gain of the hatched chicks were also followed. Residues in eggs, feces and abdominal fat were determined.

These first 13 aspects of the poultry phase were completed in July of this year and I will talk about these today.

Item 14 is being carried out at the present time and will not be discussed.

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SLIDE 2 - Our experiments consisted of 12 treatment groups with 35 hens in each group. The Aroclors are described by 4 digit numbers. The first two digits define whether it is a bi or triphenyl and the last 2 digits the % chlorine. All bi and triphenyls were fed at 20 ppm. In addition Aroclors 1242, 1248 and 1254 were fed at 2 ppm since they were the subject of the accidental contaminations. The last group of hens was fed a polybrominated biphenyl containing 75% bromine. The polybrominated biphenyl is manufactured by Michigan Chemical Co with the product name BP-6. BP-6 is a flame retardant to be used commercially in automobile seats. Not many chickens ride in automobiles, but BP-6 is a potential environmental contaminant.

The birds were fed ad libitum these compounds mixed in the feed. All treatment groups ate well and feed consumption and body weight gain were the same as the controls. There were very few deaths and no deaths could be attributed to the PCBs, PCT or polybrominated compounds.

SLIDE 3 - This slide shows eggs weight, on the bottom, and eggshell thickness on the top. The left hand side of the slide is the first 9 weeks when the hens were fed PCB. The right side of the slide is the time immediately following PCB feeding when the hens were fed clean feed. Each point is the mean of 10 eggs from each group. The line represents the control. There was no effect of any of the treatments on egg weight or eggshell thickness.



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SLIDE 4 - Although egg weight and eggshell thickness were unaffected by the treatments, most of the 20 ppm diets decreased egg production. The left hand portion of the slide shows egg production during PCB feeding. The right hand side shows the following two months when the hens were fed clean feed. Half of the data is shown at the top and half on the bottom of the slide. This slide shows the effects of feeding 20 ppm. 20 ppm 1221 and the triphenyl 5442 had no effect on egg production. All of the other treatments lowered egg production. PCB 1232, 1242, 1248 and the polybrominated biphenyl, PBB, reduced egg production to 65%. PCB 1254 and 1268 reduced egg production to a lesser extent.

The results during the no PCB feeding are a little difficult to interpret. PCB feeding started in March and the PCB feed was replaced with clean feed in May. Feeding no PCB continued until July. In late spring, with the start of warm weather, egg production normally declines. The drop in egg production in the control birds due to the increase in ambient temperature makes the interpretation difficult. However, it appears that in most of the groups egg production is returning to the expected production for the early summer season.

SLIDE 5 - This slide shows the fertility of eggs from hens fed the PCBs. The hens were inseminated with semen from control males. Each point is the mean for a treatment group with the line showing the fertility of control eggs. There was no effect of the treatments on fertility. The grey area is 85 to 95% fertility and you can see that almost all of the groups including controls were in this range.



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SLIDE 6 - Although PCBs had no effect on fertility of eggs, hatchability was reduced dramatically with the 20 ppm level of the middle members of the Aroclor series. The left hand side of the slide shows hatchability during the 9 week PCB feeding. The right hand portion of the slide shows hatchability of eggs collected during the following 6 weeks when the hens were fed clean feed. Control hatchability -- at the top of the slide -- remained about 90% throughout the experiment. Hatchability was reduced after feeding 20 ppm 1254, 1232, 1242 and 1248. The decline in hatchability started to occur after 2 weeks feeding 20 ppm. 7 weeks of feeding 1254 reduced hatchability to 70%, 7 weeks on 1232 hatchability was reduced to 40%. After 1242 and 1248 feeding very few eggs hatched. Hatchability with 1242 and 1248 feeding was only 8 to 10%.

When the PCB feed was replaced with feed containing no PCB, right hand portion of the slide -- hatchability began to improve. After 2 weeks of clean feed there was a surprising improvement in hatchability. Hatchability of hens fed 1232 and 1254 improved immediately. After 6 weeks on clean feed hatchability of all groups was better than 75%.

SLIDE 7 - As we have seen from the previous slide, 20 ppm Aroclor 1232, 1242, 1248 and 1254 reduced hatchability. Feeding these Aroclors at 2 ppm did not affect hatchability. Also 20 ppm Aroclors 1221, 1268, 5442 or the polybrominated compound BP-6 did not affect hatchability.



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SLIDE 8 - We decided to investigate the dramatic effects of the PCBs on hatchability. All unhatched eggs were examined to determine time of embryonic death and incidence of abnormalities. We were fortunate to have Dr. Jacqueline Verrett of the Food and Drug Administration collaborate with us on this phase of the study. Dr. Verrett is an expert on the effects of pesticides on embryonic development of the chick. She and her staff examined the unhatched eggs for us and this slide and the next series of slides summarize their findings. This data was only obtained recently and is not included in the preprint.

The time of embryonic death is shown on this slide. Unhatched eggs from hens fed 20 ppm 1232, 1242, 1248 and 1254 of the 5th to 9th week of feeding were combined. Most of the deaths occurred at the time of hatching. Of the 840 unhatched eggs shown, 150 died on the 20th day of incubation and 430 on the 21st day. 297 of these 430 eggs had been pipped. This means that the chick had developed to the point where it could break the shell, but was unable to break the shell completely and emerge.

SLIDE 9 - The unhatched eggs were exaimined to determine if the embryos were abnormal. Eggs from the 5th to 9th week of PCB feeding were examined. Approximately 450 eggs were set for each group. The % unhatched is an expression of hatchability. In the control group only 3% of the fertile eggs did not hatch and 2% of the fertile eggs were abnormal.

1232 --- 12% of the fertile eggs were abnormal

1242 --- 27%

1248 --- 26%

1254 --- 5% of the 450 eggs set were abnormal.

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All abnormalities were in unhatched chicks.

SLIDE 10 - The embryos were examined for embryonic defects. 843 dead embryos were examined. 286 or 34% of the 843 dead embryos were abnormal. Upon gross examination of the 286 abnormal embryos, 394 abnormalities were found. The most common abnormality observed was edema. One half of the embryos had edema. Many of the embryos had large edematous cysts on the rump and side and many had subcutaneous edema about the neck and head area. 12% of the embryos had unabsorbed yolk. A smaller percentage had small bodies or were midgets, had external hemorrhages, especially of the shell vessels. Skinny legs, rotated ankles and short or crossed beaks were also found.

Other abnormalities that occurred to a lesser extent were cleft palate, abnormal eye formations and external viscera.

SLIDE 11 - This picture is an unhatched chick after 20 days incubation. Abnormalities include a large edematous cyst on the rump and unabsorbed yolk. The large edematous cyst on the rump had been ruptured when the picture was taken.

Unfortunately 2 other slides which showed large intact edematous sacs on the rump and back of the neck were not ready in time for this talk. However, I do have prints with me if anyone wishes to see them at a later time.



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SLIDE 12 - We also determined whether the PCBs in the egg had any effect on the young that hatched. Progeny survival and growth was studied. None of the PCB treatments affected the survival of the chicks that hatched. However, growth of the chicks was impaired.

The bars to the far left of the slide, labelled C, are the body weight gains of the chicks hatched from the eggs of hens fed the control feed. A star denotes a body weight gain that is significantly less than the control. The bars next to the control are 6-9 weeks PCB. The PCBs were fed to the hens for 6 to 9 weeks, eggs collected and incubated weekly. When the chicks hatched, they were raised for 4 weeks on clean feed and the body weight gain determined. The data for the 6, 7, 8 and 9 weeks of PCB feeding were combined. Chicks that were hatched from eggs of hens fed 20 ppm Aroclor 1232, 1242, 1248 or 1254 for 6 to 9 weeks gained significantly less than the control chicks.

After 9 weeks the PCB in the diet of the hens was replaced with clean feed. Progeny studies were conducted on the eggs collected during the 4 weeks immediately after PCB feeding and is shown on the right hand portion of the slide. Eggs were collected and incubated weekly and the body weight gain of the chicks then determined. One week after PCB 1232 was removed or 2 weeks after 1254 removal from the hens diet, the chicks that hatched grew as well as the chicks from control eggs. The depletion time necessary for improved chick growth correlates well with the time necessary for hatchability recovery to occur. Aroclors 1242 and 1248 which had the greatest effect on body weight gain of the progeny, also had the longest residual effect on body weight gain. Hens had to be fed clean feed for 4 or more weeks for progeny weight gains to be comparable to controls.

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SLIDE 13 - Residue analysis of eggs and feces were done weekly. Only 10% of the PCBs fed appeared in the feces. Fecal GLC patterns were the same as the feed patterns indicating that the 10% appearing in the feces were unaltered PCBs. However, with egg and body fat samples many of the less chlorinated components were removed by metabolism. Thus the residues of Aroclor 1221 were greatly altered while the residues of Aroclor 1268 were essentially unaltered.

The analysis of the body fat and eggs of individual birds of the PCB groups are shown here. The bar graph shows the residues after feeding PCBs for 9 weeks. In general, the level of PCB in body fat at 9 weeks, increased with increasing chlorination of the formulation fed. The level in egg paralled the level in body fat. The exception was 'Aroclor 1268' where the level in eggs was relatively higher than expected. The excretion of 1268 in the eggs could explain the low accumulation of 1268 in the fat.

The bottom of the slide shows depletion of residues after 6 weeks on clean feed. Aroclor 1242 appears to be removed more easily than any of the other Aroclors .

The depletion of residues after feeding stops depends on metabolism, level of egg production and changes in body composition. While depletion occurred with all compounds, a thorough analysis of all factors has not been made.

There is poor correlation of the amount of residue in the egg with the teratogenic effects. PCBs 1242 and 1248 which had the greatest adverse effects did not have higher egg residues than 1254 or 1268. In addition 1232 had a very small accumulation in the egg - only 3 ppm - but its effects on hatchability and embryonic development were greater than 1254's effects.



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At the present time the exact component which is responsible for the deleterious effects of specific Aroclors is unknown.

LAST SLIDE PLEASE

In summary:

A. PCBs fed to White Leghorn Laying Hens

No effect on:

Feed consumption & body weight gain

Mortality

Egg weight & eggshell thickness

Fertility

B. 20 ppm 'Aroclor' 1232, 1242, 1248 and 1254

Reduced egg production

Reduced hatchability

Caused teratogenic effects

Reduced body weight gain of progeny

C. PCB Residues

Accumulate in egg at 1/2 to 1/10 dietary level

Fat residue = 10 x egg

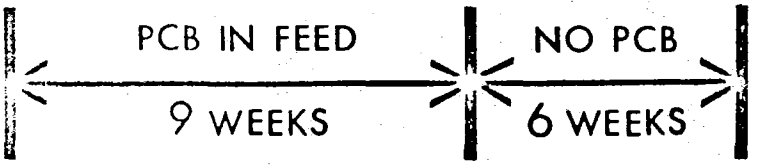
Depletion not complete after 6 weeks feeding of clean feed.



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## EXPERIMENTAL DESIGN

MATURE WHITE LEGHORN HENS FED ad libitum




1. FEED CONSUMPTION
2. BODY WEIGHT GAIN
3. MORTALITY
4. EGG WEIGHT
5. EGGSHELL THICKNESS
6. EGG PRODUCTION
7. FERTILITY
8. HATCHABILITY
9. TERATOGENIC EFFECTS
10. SURVIVAL OF PROGENY
11. RESIDUES IN EGGS
12. RESIDUES IN FECES
13. RESIDUES IN ABDOMINAL FAT
14. DRIED MANURE RECYCLED, IN DAIRY COWS
    a. RESIDUES IN MILK
    b. RESIDUES IN BODY FAT
    c. DEPLETION OF RESIDUES

SLIDE 1.

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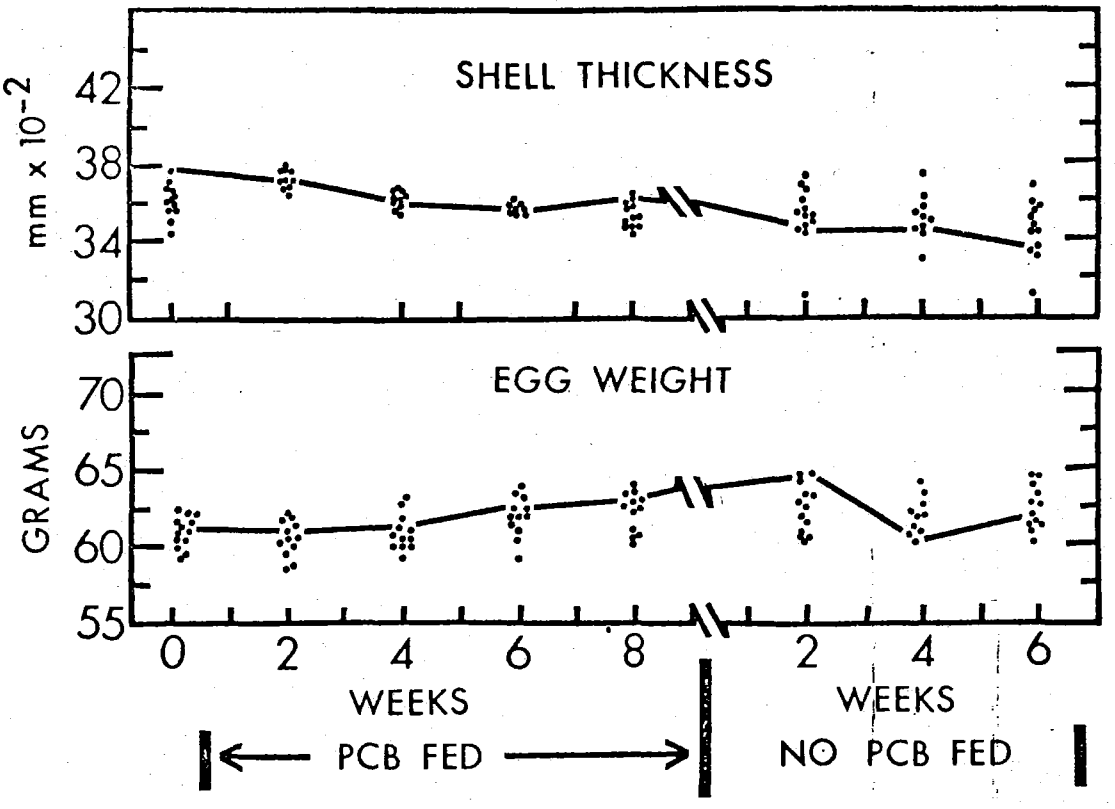
TREATMENTS

12 GROUPS WITH 35 HENS PER GROUP

| | | | | |
|---|---|---|---|---|
| CONTROL | | | | |
| 20 ppm | 1221 | PCB | 21% Cl | (AROCLORS MONSANTO) |
| 20 ppm | 1232 | PCB | 32% Cl | |
| 2 ppm<br>20 ppm | 1242<br>1242 | PCB | 42% Cl | |
| 2 ppm<br>20 ppm | 1248<br>1248 | PCB | 48% Cl | |
| 2 ppm<br>20 ppm | 1254<br>1254 | PCB | 54% Cl | |
| 20 ppm | 1268 | PCB | 68% Cl | |
| 20 ppm | 5442 | PCT | 42% Cl | |
| 20 ppm | BP-6 | PBB | 75% Br | (BP-6 MICHIGAN) |

SLIDE 2

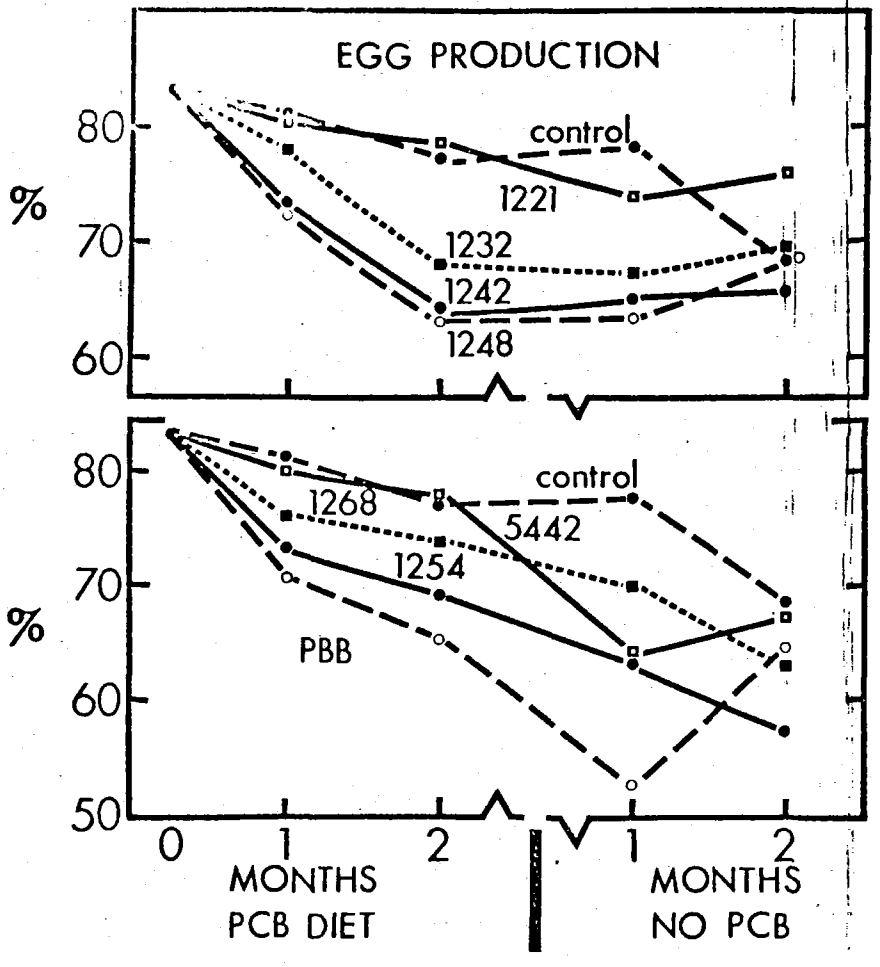
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SLIDE 3

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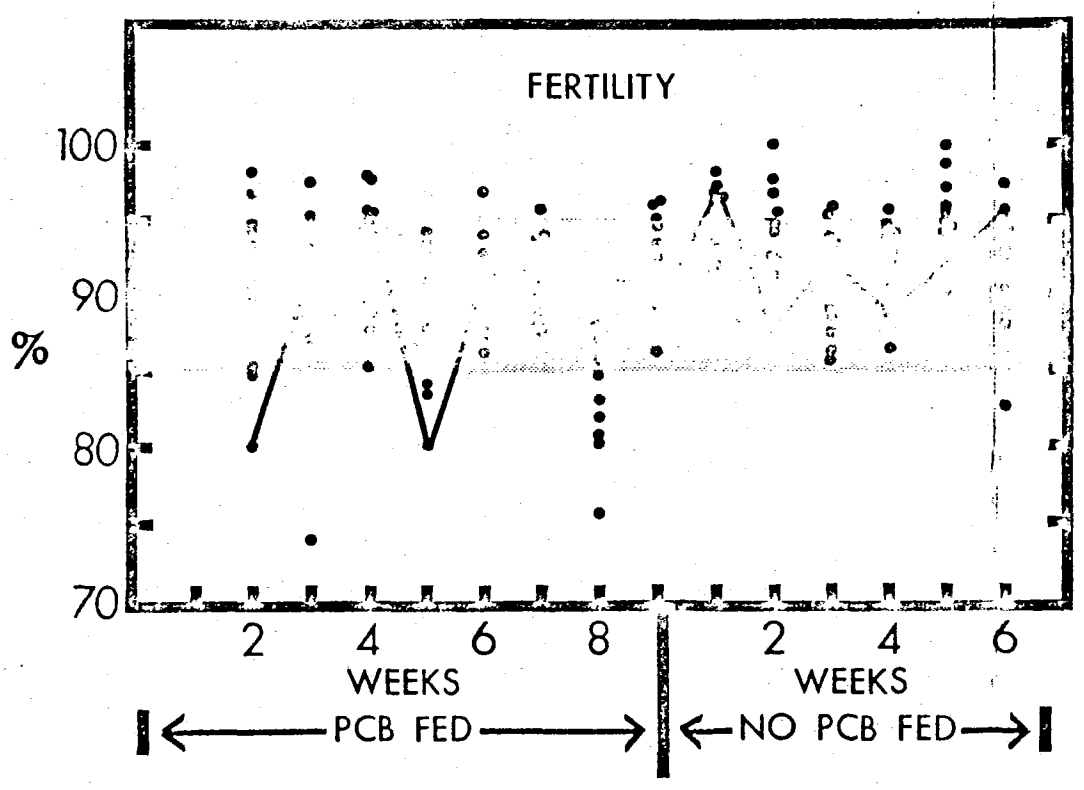
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SLIDE 4

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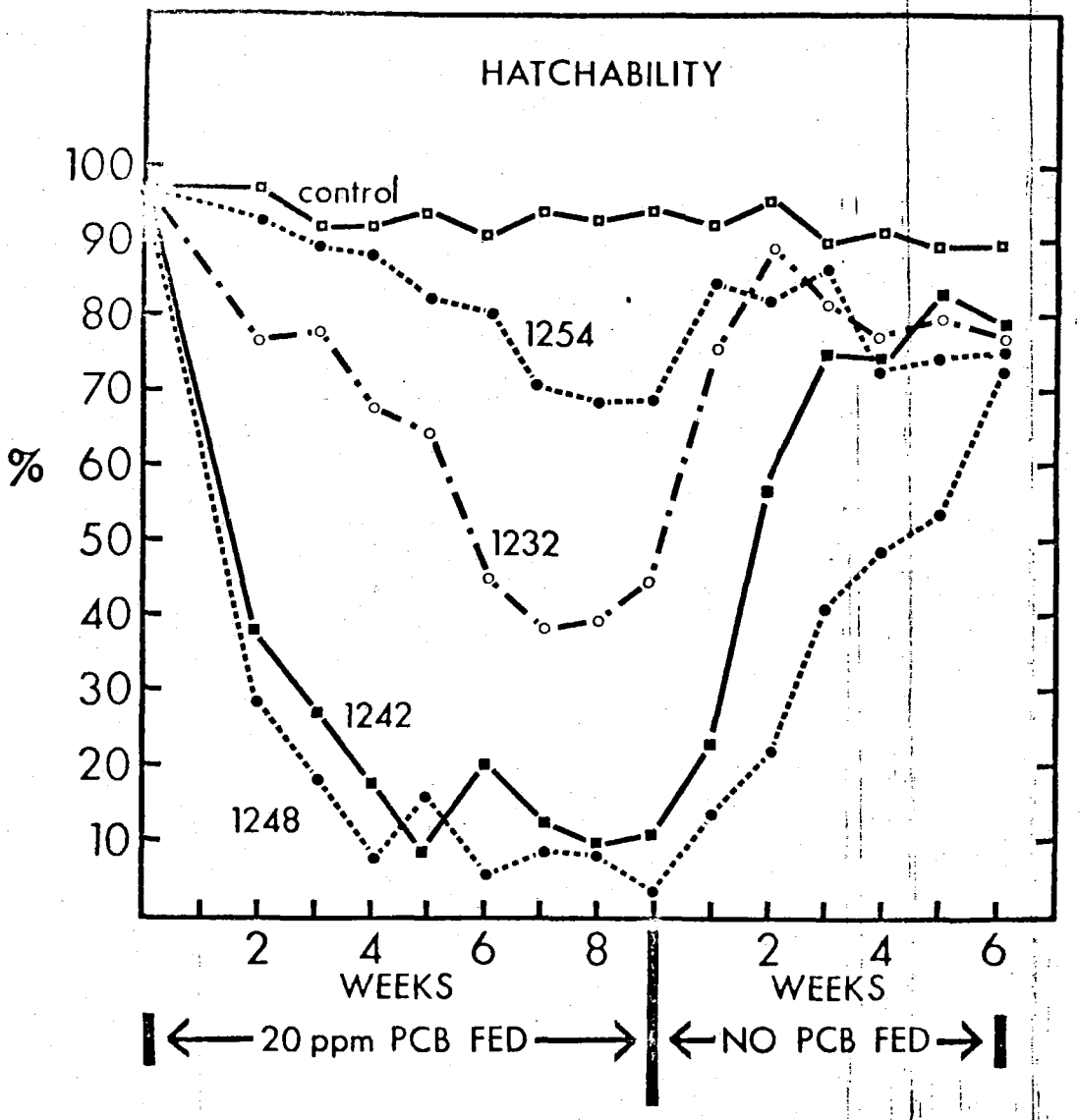
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SLIDE 5

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SLIDE 6

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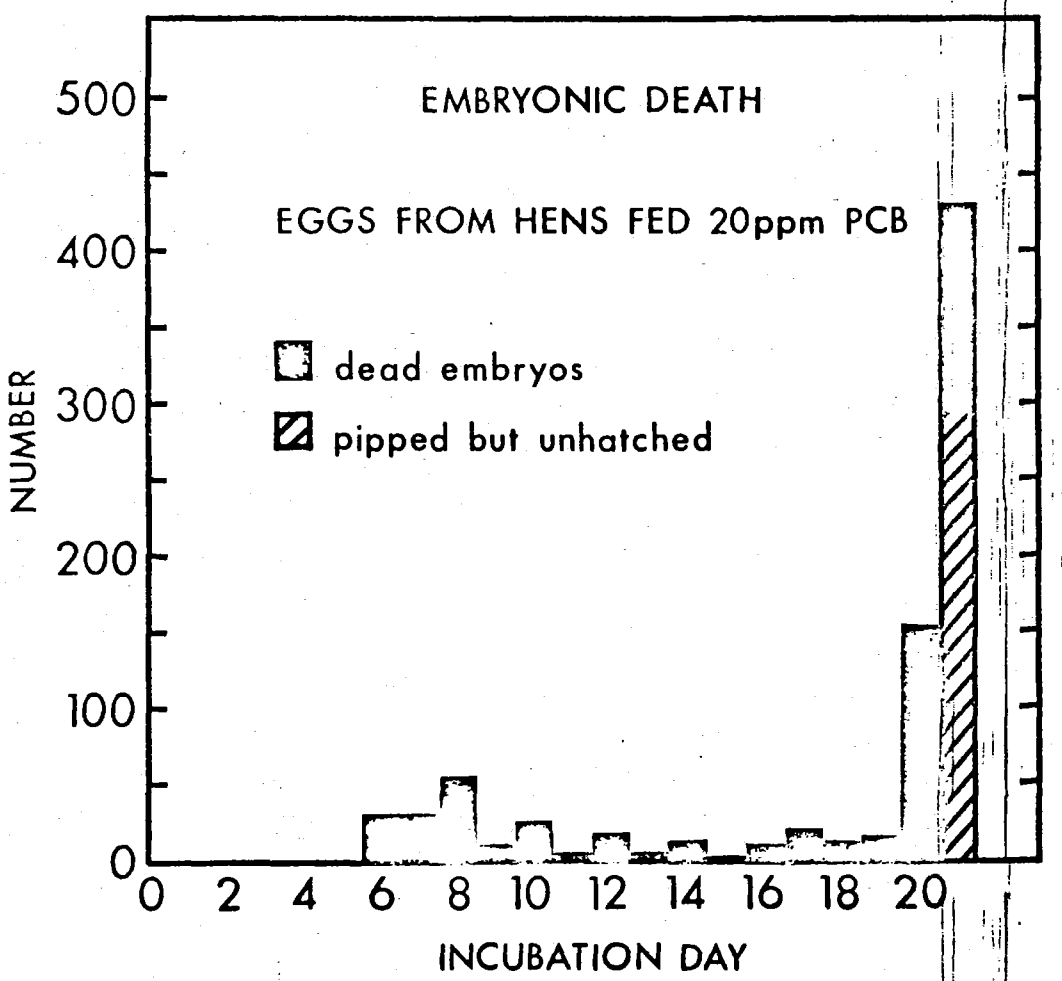
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| DECREASED HATCHABILITY | |
|---|---|
| 20 ppm | 1232 |
| | 1242 |
| | 1248 |
| | 1254 |
| **NO EFFECT ON HATCHABILITY** | |
| 2 ppm | 1232,1242,1248,1254 |
| 20 ppm | 1221 |
| | 1268 |
| | 5442 (PCT) |
| | PBB-6 |

SLIDE 7

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SLIDE 8

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## EMBRYONIC DEATH & ABNORMALITIES

| | % Unhatched | % Abnormal |
|---|---|---|
| Control | 3 | 2 |
| 20 ppm 1232 | 39 | 12 |
| 20 ppm 1242 | 75 | 27 |
| 20 ppm 1248 | 76 | 26 |
| 20 ppm 1254 | 18 | 5 |

SLIDE 9

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## ABNORMALITIES IN DEAD EMBRYOS

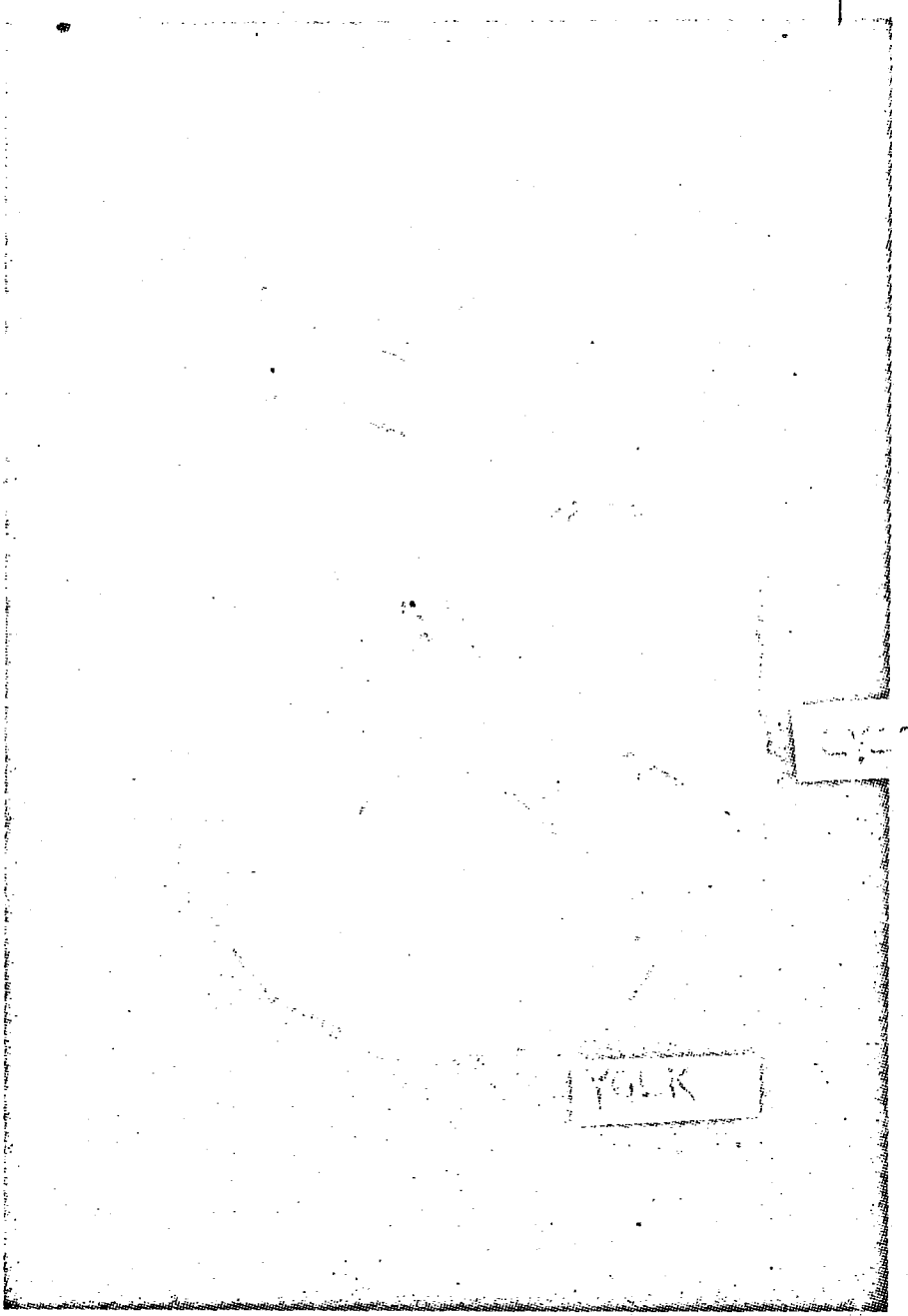
20 ppm 1232, 1242, 1248 OR 1254

| | NUMBER | % |
|---|---|---|
| DEAD EMBRYOS | 843 | 100 |
| ABNORMAL EMBRYOS | 286 | 34 |
| | | |
| ABNORMALITIES | 394 | 100 |
| EDEMA | 198 | 50 |
| UNABSORBED YOLK | 49 | 12 |
| SMALL BODY | 34 | 9 |
| EXTERNAL HEMORRHAGE | 30 | 8 |
| SKINNY LEGS | 28 | 7 |
| ROTATED ANKLE | 12 | 3 |
| SHORT or CROSSED BEAK | 11 | 3 |
| OTHERS | 32 | 8 |

SLIDE 10

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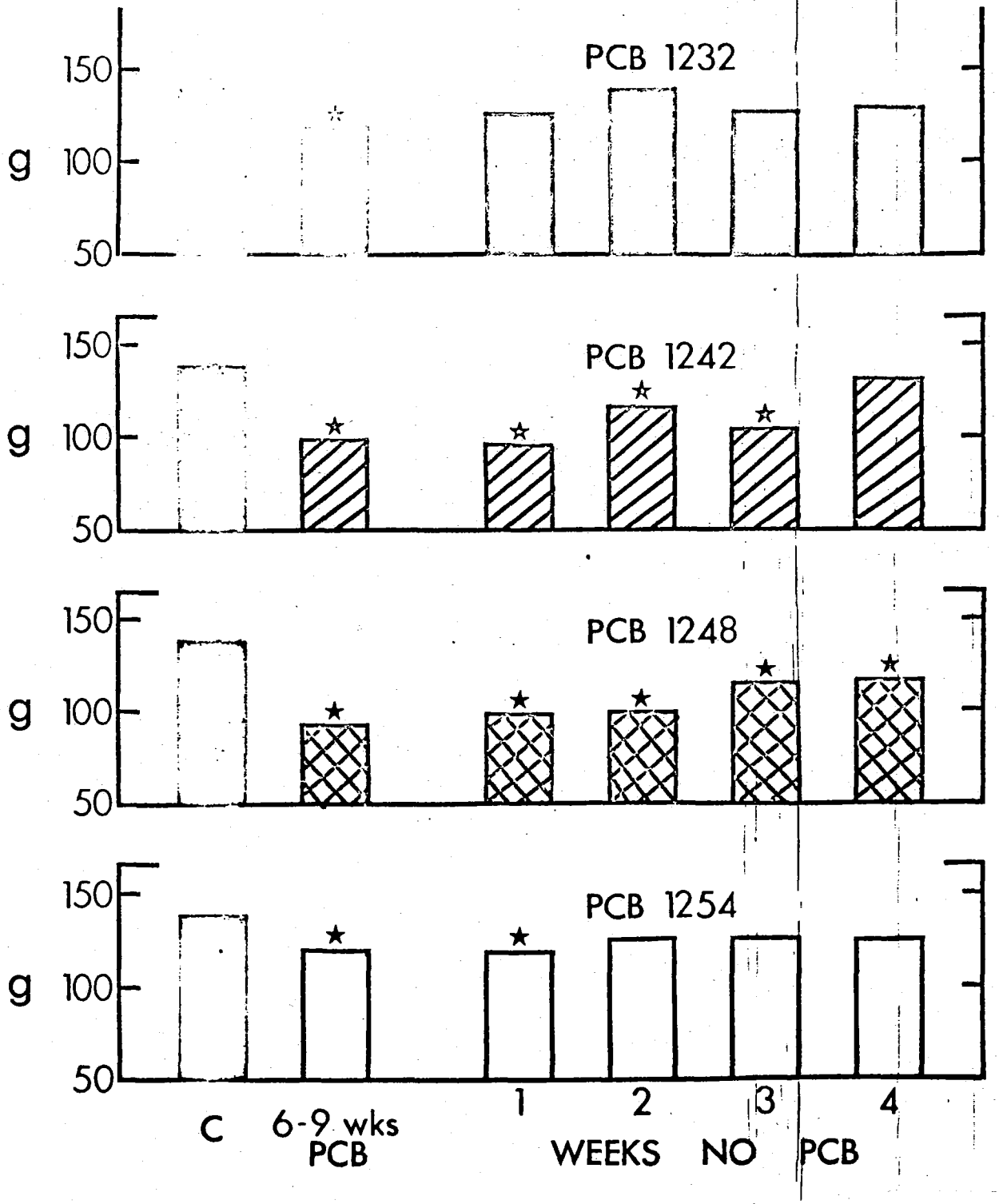
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SLIDE 11

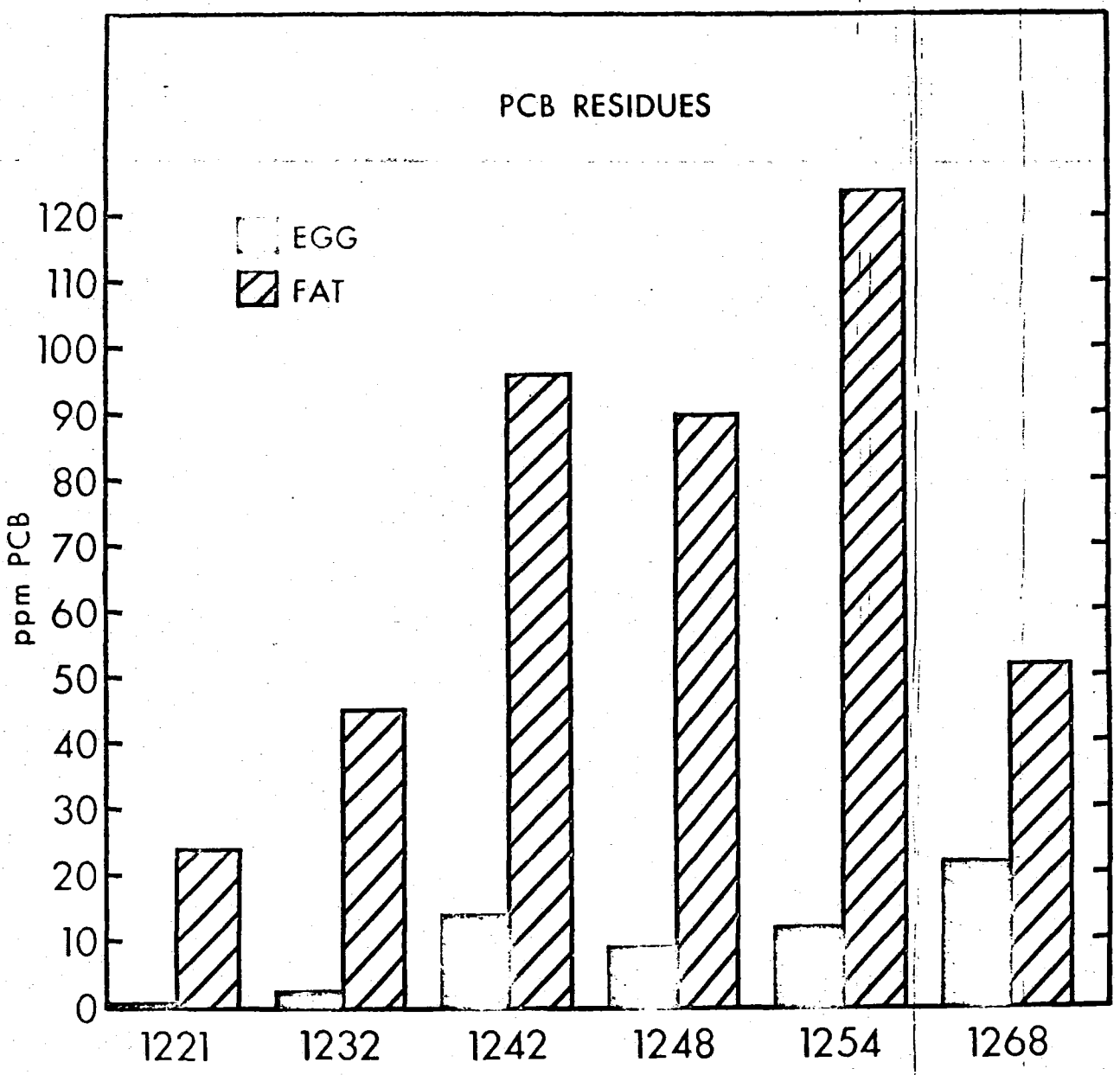
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BODY WEIGHT GAIN OF CHICKS HATCHED FROM EGGS OF HENS FED PCB




SLIDE 12

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% DEPLETION:

| | | | | | | |
|---|---|---|---|---|---|---|
| EGG | 0 | 17 | 80 | 67 | 42 | 92 |
| FAT | 66 | 48 | 93 | 56 | 26 | 17 |

SLIDE 13

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## SUMMARY

A. PCBs FED TO WHITE LEGHORN LAYING HENS

NO EFFECT ON:

- FEED CONSUMPTION & BODY WEIGHT GAIN
- MORTALITY
- EGG WEIGHT & EGGSHELL THICKNESS
- FERTILITY

B. 20ppm PCB 1232, 1242, 1248, 1254

- REDUCED EGG PRODUCTION
- REDUCED HATCHABILITY
- CAUSED TERATOGENIC EFFECTS
- REDUCED BODY WEIGHT GAIN OF PROGENY

C. PCB RESIDUES

- ACCUMILATE IN EGG AT $\frac{1}{2}$ TO $\frac{1}{10}$ DIETARY LEVEL
- FAT RESIDUE = 10 x EGG
- DEPLETION NOT COMPLETE AFTER 6 WEEKS

SLIDE 14

DSW 369652

*From the desk of*
W. B. PAPAGEORGE

8/14

J. F. Stapleton:

Attached is a copy of the FDA report "Survey: Polychlorinated Biphenyls in Milk" referred to in the FDA report received earlier "PCBs in Milk Survey Data", paragraph 5, dated May 17, 1972

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