# Some Observations Concerning the Ratio of the Intake of Organochlorine Insecticides Through Food and Amounts Excreted in the Milk of Breast-feeding Mothers

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In earlier studies Ann., (1969); ADAMOVIĆ et al. (1971) it was established that significant amounts of organochlorine insecticides, especially DDT derivatives, were present in human milk. Compared with cows milk, human milk contained 20 times the amount of the insecticide Ann., (1969); ADAMOVIĆ et al. (1970). When comparing the daily intake of DDT derivatives through food and the daily excreted amounts of the insecticide through human milk, a "surplus" amount of the insecticide excreted is revealed ADAMOVIĆ et al. (1971, 1971a). It is assumed that this "surplus" arises due to the mobilized concentration of the DDT derivative, especially pp<sup>2</sup> DDE, present in the mother body ADAMOVIĆ and SOKIĆ (1973).

This study represents a continuation of our recent works ADAMOVIĆ et al. (1971, 1971a), carried out under more organized conditions with the aim of verifying the earlier acquired data and rendering the required corrections.

### Materials and Methods

The work was conducted in cooperation with the Special Hospital for Premature Children in Belgrade. Two groups, each consisting of five breast-feeding mothers, were selected. Group A consisted of mothers feeding one infant and group B was feeding twins. During the preparatory work for the research special attention was given to the selection of the participants.

This, thus provided a fairly homogenous structure of the groups:

- of similar group (26 3);
- city inhabitants;
- not engaged in professions which might affect the results;
- have similar previous experiences in number of childbirths (first childbirths experience for all) and abortions (only two women);

- that they were expecting a child at about the same time (30 days prior to childbirth);
- that they had normal eating habits;

In addition to this, the total amount of excreted milk was measured every day, as was body weight at the beginning and the end of the seven day experiment.

Both groups of participants received the same quantity and content of daily meals. During the experiment they were not given additional foods and also were not taking anticionvulsives on the basis of phenobarbital or diphenylhydantion DAVIS et al. (1969); KWALICK (1971).

The experiment lasted for seven consecutive days during which time the examination of some complete daily meals (breakfast, lunch, dinner, between-meals snacks, fruit and bread) commenced a day defore the human milk was examined. A total of seven daily meals was examined.

The milk of each mother was taken 5 times during the course of 24 hours. The test sample was the average of that quantity taken from each individual mother; a total of 70 samples of human milk were examined.

The meals are first roughly homogenized and then with the aid of the Sorwal homogenizer further made into a fine paste. The aliquot part is mixed with anhydrous sodium sulphate and dried overnight at a temperature of  $40^{\circ}$ C. Further, it was extracted with a highly purified petrol ether in a Soxhlet apparatus for 24 hours. The crude fat is cleaned up by extraction with acetonitrile and petrol ether and thenthrough a column of deactivated Florisil and elutes into two fractions: petrol ether and 5% ethyl ether in petrol ether, ADA MOVIĆ. The aluates are concentrated to the required volume and then analyzed by means of gas-liquid chromatography.

The samples of milk are deproteinized by ethanol, centrifuged and then extracted with ethyl ether and twice with petrol ether. The extracts are combined, evaporated to dryness, the residue dissolved in petrol ether and subsequently purified as was the crude fat meal.

The instruments used were Varian Models 1440 and 2740 with electron capture detectors  $(H_3)$  and Pyrex glass columns: 6' x 1/8" filled with a mixture 1.5% OV-17 and 1.9% QF-1 (1:1) on Gas Chrom Q and 10% DC-200 and 15% QF-1 (1:1) on Gas Chrom Q, 100-200 mesh; the carrier gas was prepurified nitrogen.

Identification is carried out by comparison of the pure insecticide substances under the same conditions as well as measuring

the p-value. The value is determined by measuring the peak height; recovery amounted to 85-102%. Data given in Table 1 and 2 was not corrected for recovery.

#### Results and Discussion

In our recent studies ADAMOVIĆ et al. (1971, 1971a) it was established that a greater amount of DDT derivatives was excreted through human milk than is daily taken in by food - Table 1 and 2. Perticularly evident is the disproportion between the intake of pp'DDE and the excreted amount, which is not really the case with other derivatives of DDT. This phenomenon of "surplus" was not observed to occur with other insecticides (alpha-, beta-, gamma-, BHC, dieldrin) present in the tested meals i.e. in human milk.

The second phenomenon observed, also in our studies of 1971, pointed to the significant increase in the excretion of this metabolite in the milk of breast-feeding mothers with twins (B) in comparison to the milk of mothers breast-feeding one child (A): 0.599 ppm and 0.126 ppm - Table 1. If by comparing the daily excreted amounts of pp'DDE via milk and amounts taken through food in both groups of participants, especially considering the daily excreted average quantities of milk, then these differences are particularly evident: mothers of twins (B) excrete 12 times more the amount of pp'DDE i.e. 4 times more than was the total as DDT; while with mothers having only one child (A) the condition is the opposite - they excrete only about 40% of the total as DDT taken in daily through food - Table 2.

The defferences were far too great in order that we should accept the possible conclusions following from the above data. Firstly, it was necessary for us to repeat the experiment and to minimize some of the observed shortcomings, dominant amongst which was the lack in homogeneity in and among the group participants (age, number of childbirths, time of lactation and so on), Apart from other things the average amounts of milk excreted daily were fairly similar in quantity, in contrast to the prior case: 854 ml  $(A_1)$  and 950 ml  $(B_1)$  - Table 2.

Thus, in contrast to the prior data, where mothers of twins (B) excreted daily in their milk 12 times more pp³DDE than that of mothers with one child (A) - Table 2, now data obtained for both groups was similar: Group A<sub>1</sub> excreted 13 times a greater amount of pp³DDE (161 gamma/day), i.e. 3.7 times more than the total as DDT (223 gamma/day) and group B<sub>1</sub> only 11 times greater amount (135 gamma/day) i.e., 3 times more (187 gamma/day). Taking into account the high level of homogeneity achieved in this work, we are inclined to correct the earlier, provisional conclusion concerning the increased excretion of the DDT derivative by breast-feeding

mothers with twins. This described phenomenon is present thereby in all breast-feeding mothers, regardless of number of infants. We are of the opinion that during the process of lactation a powerful mobilizing process of the deposited DDT derivatives occurs, aspecially of pp'DDE which is then excreted through milk. The high ratio existing between DDE/DDT which was on average 78% in the examined human milk, also supports these opinions. The same ratio was 68% in the abdominal fatty tissue, the brain, adrenal tissue and other human organs found present in the general population of Serbia.

TABLE 1

A Comparative Representation of the Content of DDT Derivates and Dieldrin in the Milk of Breast-feeding Mothers with One Infant or Twins and Their Presence in all the Daily Meals during the Years 1971 and 1974 (expressed in ppm)

Insec- ticide	1971			1974		
	A	В	total daily meal	A 1	В <sub>1</sub>	total daily meal
pp' DDE	0,126	0.559	0.024	0.192	0.142	0.005
pp' DDD	0.006	0.010	0.008	tr.	tr.	0.003
pp' DDT	0.048	0.120	0.042	0.048	0.036	0.011
op' DDT	0.003	0.006	0.023	0.003	0.004	0.004
total as DDT	0.198	0.752	0.095	0.264	0.197	0.024
dieldrin	0.001	0.004	0,003	0.001	tr.	0.001

A and A - milk of breast-feeding mothers with one child B and B - milk of breast-feeding mothers with twins tr < 0,001 ppm

These explanations concerinng presence of the "surplus" of the DDT derivative in human milk are also based on the assumption that food is the source of 95% of the taken-in and deposited pesticides. However, if it is established that food is responsible for only 50% of the present pesticides in the human body and that the other 50% results from still-inconclusively defined sources, such as air, water, dust, tobacco and cosmetics Ann., (1969); EDMUNDSON et al. (1967) then the above given data will be appropriately corrected. Even then, however, a "surplus" would exist which would require an explanation. Errors which would result from the individual use of cosmetics and tobacco could not be avoided in this study.

TABLE 2

A Comparative Representation of the Average Daily Excretion through Milk and the Average Daily Intake through Food of DDT Derivates and Dieldrin in Mothers with One Infant and Twins during the years 1971 and 1974 (expressed in gamma/total daily excreted amount of milk i.e. intake of food)

	1971			1974		
Insec- ticide	A	В	Dail <b>y</b> intake through food	A <sub>1</sub>	B <sub>1</sub>	Daily intake through food
pp'DDE	67	787	65	161	135	12
pp'DDD	3	15	20	tr.	tr.	7
pp'DDT	25	167	112	41	34	29
op'DDT	2	9	59	2.8	3.3	11
Total as DDT	105	1.056	266	223	187	60
%DDE/ /DDT	71	83	27	80	80	22
dieldrin	tr.	10.6	9	1	tr.	2
Average weight of milk % fats	527 ml 3.8	1.420 ml 3.2	2.659 g 5.8	854 ml 4.1	950 ml 3.7	2.478 g. 5.2

A and A - breast-feeding mothers with one child B and B - breast-feeding mothers with twins tr.  ${<0.001}$  ppm.

It is interesting to note, apart from these observations, that even following reductions in amounts of intake of the DDT derivative by 75% through food (Table 1), achieved in the last three years by adhering to restrictive measures and other regulations in Yugoslavia, the level of this pesticide in human milk has not reduced significantly:

Pesticide Expressed as DDT Present in Human Milk, in ppm

1967	0.207, ADAMOVIĆ et al. (1970a)
1968	0.321, ADAMOVIĆ et al. (1970)
1971	0.198, and 0.752 ADAMOVIĆ et al.
	(1971; 1971a)
1974	0.264 and 0.197 (Table 1)

This data indicates that a newborn child has a daily intake of 200 gamma of total as DDT through mother's milk. This represents a 100% increase, in relation to the daily intake of the adult population of Serbia ADAMOVIĆ et al. (1975). The risk for the youngest generation is evident, especially when the upper limit of the tolerated daily intake of pesticides for adults is: 350 gamma/day/person i.e. the recommenden intake for children is 1/10 of this dosage FAO//WHO (1972).

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