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Evaluation of Infant Formula Contamination in Italy

C. Finoli & G. Rondinini

Dipartimento di Scienze e Tecnologie Alimentari e Microbiologiche,
Universita' degli Studi di Milano,
Via G. Celoria, 2, 20133 Milano, Italy

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ABSTRACT

Twenty-six liquid and powdered milk infant formulas have been subjected to tests for moisture or dry matter, ash, trace elements, filth, lipopolysaccharide, and aflatoxin M_1 content and microbial evaluation.

Microbial counts are lower than the Italian legal limit except for two powder samples. On the other hand, the lipopolysaccharide content, an indirect index of gram negative contamination, indicates in this respect, poor raw material quality.

Only three powdered milk samples contain aflatoxin M_1, the concentration of which does not exceed 13 ppt in the reconstituted milk. Pb levels do not exceed 0·25 ppm.

All the results give a satisfactory hygienic picture for these products, but there is room for improvement.

INTRODUCTION

According to Italian law, infant formulas are those foods that completely or partially substitute breast-feeding and are used for weaning or for the alimentary integration of infants during the early period of life (*Gazzetta Ufficiale*, 1951).

We have already reported on the chlorinated pesticide residues, trace elements, filth, and microbial evaluation that comprise the contamination of human milk and infant formulas (Cerutti *et al.*, 1978; Finoli *et al.*, 1978;

Food Chemistry 0308-8146/89/$03·50

Ottogalli *et al.*, 1982). We now want to consider, in addition to some of the above cited parameters, the lipopolysaccharide (LPS) content, an indirect index of gram negative contamination, and the aflatoxin M_1 (AFM_1) concentration, whose presence in Italian infant formulas has already been reported. (Riberzani *et al.*, 1983; Quintavalla & Casolari, 1985; Vittani, 1987).

MATERIALS AND METHODS

Eight kinds of ready to use liquid milk (normally used in the pediatric departments of some hospitals of Milan), from five firms marked as A, B, C, D, and E and 18 kinds of powdered milk infant formulas, from six different manufacturers marked as B, C, D, E, F, and G and commonly found in chemists' shops, were examined for:

—moisture or dry matter (Federation Internationale de Laiterie, 1964; 1982)
—ash (Cerutti *et al.*, 1978)
—aflatoxin M_1 (Vittani, 1987)
—trace elements (Cerutti *et al.*, 1978)
—filth (Ottogalli *et al.*, 1982)
—lipopolysaccharides by LAL test (Rondinini *et al.*, 1984; 1985).

All samples were also submitted to microbial analysis according to Italian law (*Gazzetta Ufficiale*, 1978).

On the powdered milks the following determinations were also carried out:

—thermoduric microorganisms, in Plate Count Agar (Difco), pour plate technique, after sample pasteurization at 63°C for 30 min, incubation at 32°C for 48–72 h.
—sulphite-reducing clostridia, in Sulphite Polymixin Sulphadiazine Agar (Biolife), inoculated tubes sealed with sterilized paraffin, incubation at 35°C under anaerobic conditions for 48–72 h.
—butyric clostridia, according to Weinzirl (1921).

Reconstitution of the powdered milk was carried out according to the firms' instructions with a dilution ratio ranging from 5·7 to 7·6.

RESULTS

The results of moisture or dry matter, ash and trace elements are reported in Tables 1 and 2.

AFM_1 concentration is below the method detection limit, i.e. 3 ppt, in all

TABLE 1
Liquid Milk: Dry Matter, Ash, and Trace Elements

Manufacturer	*Samples*	*Dry matter* (%)	*Ash* (%)	*Fe*	*Cu*	*Zn*	*Mn*	*Co* (*ppm*)	*Cr*	*Ni*	*Pb*	*Cd*
A												
	1[a]	13·43	0·43	10·25	0·35	2·60	0·05	0·08	0·06	0·04	0·05	0·10
	2[a]	14·18	0·27	6·75	0·22	3·15	0·12	<0·05	<0·05	0·04	0·05	<0·02
	3[a]	15·34	0·66	1·85	0·05	2·18	0·12	0·18	<0·05	0·18	0·20	0·02
B												
	4[b]	12·92	0·33	6·25	0·38	2·40	0·05	<0·05	0·05	0·06	<0·05	<0·02
	5[b]	12·81	0·35	5·50	0·40	4·85	0·06	<0·05	0·06	0·06	<0·05	<0·02
C												
	6[b]	13·84	0·35	8·75	0·40	4·25	0·12	0·05	0·08	0·08	0·05	<0·02
D												
	7[b]	11·46	0·30	6·00	0·40	2·42	0·06	<0·05	0·05	0·05	<0·05	<0·02
E												
	8[b]	13·58	0·40	1·55	0·55	5·25	0·36	0·08	0·08	0·09	0·10	<0·02

[a] Bottle.
[b] Can.

TABLE 2
Powdered Milk: Moisture, Ash, and Trace Elements

Manufacturer	*Samples*	*Moisture* (%)	*Ash* (%)	*Fe*	*Cu*	*Zn*	*Mn*	*Co* (*ppm*)	*Cr*	*Ni*	*Pb*	*Cd*
B												
	1	2·47	2·36	49·5	2·10	29·0	0·75	1·05	0·55	0·50	0·90	<0·05
	2	2·42	3·64	100	2·40	29·5	0·50	1·30	<0·10	1·45	1·35	<0·05
	3	3·27	3·76	85·0	2·20	31·0	2·50	1·80	0·70	0·60	1·40	0·15
	4	2·62	3·42	87·5	3·70	38·0	0·50	1·25	<0·10	0·70	1·55	<0·05
	5	2·25	2·39	60·0	2·75	30·0	1·00	0·75	<0·10	0·60	1·10	<0·05
	6	2·75	2·33	67·5	3·90	31·5	0·75	<0·10	2·10	1·75	0·75	<0·05
C												
	7	2·70	2·34	77·5	3·00	24·5	1·00	0·70	<0·10	0·65	0·75	<0·05
	8	3·02	4·07	60·0	2·80	22·7	0·95	1·25	<0·10	0·60	0·60	<0·05
D												
	9	2·48	3·04	17·8	0·50	8·75	0·50	<0·10	0·35	0·60	0·50	<0·05
	10	2·58	2·13	17·5	0·35	8·25	<0·10	<0·10	0·50	0·75	0·60	<0·05
	11	2·29	2·08	95·0	0·35	8·50	<0·10	<0·10	0·75	1·00	0·80	<0·05
	12	3·23	3·09	60·0	0·75	12·0	0·75	0·90	<0·10	0·95	0·75	0·10
	13	2·67	2·41	72·5	2·75	20·3	4·75	0·85	<0·10	1·25	0·90	0·10
	14	3·49	5·32	10·8	1·00	15·8	3·00	1·40	<0·10	1·45	1·00	0·20
E												
	15	1·54	2·78	15·0	3·80	40·8	<0·10	<0·10	0·80	1·25	0·50	<0·05
	16	2·15	3·41	90·0	4·15	38·3	2·75	<0·10	1·10	1·50	0·95	<0·05
F												
	17	1·26	2·35	49·0	2·70	23·5	1·00	<0·10	1·60	1·75	0·65	<0·05
G												
	18	2·21	1·71	6·50	0·95	6·10	<0·10	<0·10	1·25	1·00	0·35	<0·05

liquid and in 15 out of the 18 powdered samples. In the three contaminated samples, labelled as 2B-9D-14D, the toxin concentration is 64, 40, and 95 ng/kg, respectively; that is, 10, 6, and 13 ng/litre in the reconstituted milks.

Non-microbial contaminants, such as insect fragments, are not found in any samples. On the other hand, synthetic fibres are always present, ranging from 2 to 5 and from 2 to 48 in 500 ml of liquid or reconstituted powdered milks, respectively. In the liquid milks there are also some vegetal fibres, animal hair, metal and mineral fragments in 2, 3, 1, and 5 samples respectively. In the powered milks vegetal fibres and animal hair fragments are present in four, and metal fragments in three samples.

LPS content ranges from 50 to 1330 ng/ml for liquid samples and from 50 to 10 500 ng/g for powdered milks. The indirect evaluation of gram negative contamination, carried out according to Rondinini *et al.* (1984), who state 1 ng LPS to be equivalent to 4000 gram negative CFU, shows values ranging from 2×10^5 to $5{\cdot}3 \times 10^6$ CFU/ml for liquid samples and from 2×10^5 to $4{\cdot}2 \times 10^7$ CFU/g for powdered milks.

In the powdered milk the aerobic mesophilic flora is always below 200 CFU/g; thermodurics, coliforms, sulphite-reducing clostridia, and salmonella are always absent. *Staphilococcus aureus* is present in two samples from different manufacturers (8C, 18G), and butyric clostridia are present in two samples of the same firm (11D, 14D).

The microbial analysis on liquid milks shows a constant absence of viable microflora.

DISCUSSION AND CONCLUSION

Dry matter, moisture and ash levels are normal for these kinds of products and agree with the firms' statements.

Ordinary microbial analysis shows values much lower than the Italian limits (*Gazzetta Ufficiale*, 1978) except for the presence of *Staphylococcus aureus* in two samples; our values agree with those of Rivas *et al.* (1985), Umoh *et al.* (1985), Alvarez Marante *et al.* (1986), Cullen *et al.* (1986). Contrary to Rowe *et al.* (1987) findings in some English infant formulas, *Salmonella* is not found.

On the other hand, the LAL test shows poor quality of raw materials, restricted to indirect gram negative evaluation. This shows how useful the LAL test is to establish the original microbial quality of heat-treated foods.

The filth test shows that the tested milks are of a good quality; in fact, insect fragments are absent and metal fragments and animal hairs are scantily present. Numerically significant are the synthetic fibres, the results of generic environmental pollution.

AFM_1, when present, shows modest concentrations of the same magnitude as those reported by Vittani (1987) and much lower than those of Riberzani *et al.* (1983) and of Quintavalla & Casolari (1985). Only one sample, showing 13 ppt in the reconstituted milk, exceeds the Swiss legal limit, i.e. 10 ppt (*Raccolta Sistematica*, 1981), which is one of the strictest in these matters.

This limit should be adopted by Italian law, which is presently lacking in this regard; in fact, as far as mycotoxins in Italy are concerned, only the AFB_1 in feedstuffs (*Gazzetta Ufficiale*, 1976) is regulated.

Trace element concentrations in liquid and powdered milk, when diluted, are of the same magnitude as those advertised on the labels. The observed concentrations of Fe, Cu, Zn, and Mn agree with the literature and satisfy the normal nutritional requirements of infants. Although some elements seem to exceed FAO/WHO (XXVII Report) guidelines, it should be considered that the assimilation of these added elements is different in respect to that of the same naturally present in human milk. Co levels are higher than those observed by Cerutti *et al.* (1978) in human milk. Cr and Ni, whose deficiency provokes some pathological consequences, are present at concentrations much lower than those which can induce toxicological phenomena. Cd, lower than method sensitivity in 77% of the samples, shows a maximum value of 0·2 ppm in only one sample. Calculating for the weekly consumption of this powdered milk, the Cd ingestion is about one-third of that considered tolerable by the FAO/WHO (XVI Report). Pb is practically absent in three samples and ranges, when powder dilution is accounted for, from 0·05 to 0·25 ppm in the others. For this element the FAO/WHO (XXII Report) considers a weekly intake not exceeding 3 mg as acceptable for adults; FAO/WHO underlines that it is impossible to state a Pb limit for children, who belong, along with pregnant women, to the most sensitive category. The observed Pb levels are of the same magnitude as those reported by Pavelka *et al.* (1985), Tseng (1985), Khalid *et al.* (1987) but higher than those of Dabeka & McKenzie (1987).

In conclusion, the results show a satisfactory hygienic picture for these products. However, we emphasize, considering the consumers of these kinds of products, that firms should pay more attention to raw material quality in respect to gram negative contamination, as well as complete avoidance of the presence of AFM_1 and heavy metals.

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