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Evaluation of Polyethylene Intramammary Device (IMD) in Mastitis Control

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EVALUATION OF POLYETHYLENE INTRAMAMMARY DEVICE (IMD) IN
MASTITIS CONTROL

M. J. Paape, G. Ziv, and W. D. Schultze

Abstract

An abraded intramammary device (IMD) was developed. When inserted into the mammary gland cistern, the abraded IMD increased the milk somatic cell count (MSCC) in stripping milk to approximately 900,000/ml. The MSCC in the main milk remained low, averaging approximately 70,000/ml. Experimental challenge studies indicated that the abraded IMD was approximately 70% effective in the prevention of infection after experimental challenge with Staphylococcus aureus, Streptococcus uberis and Escherichia coli. It was observed, however, that within a year the abraded IMD became coated with an amorphous material, with a concomitant decrease in stripping MSCC to concentrations that were no longer effective in preventing infection. Coating the abraded IMD with sulfur hexafluoride appeared to prevent this build-up of amorphous material.

Results from field testing of the abraded IMD indicate that the IMD was highly successful in reducing the incidence of clinical mastitis. Thirteen dairy herds totaling 5000 cows were selected for study. Abraded IMD were inserted into all four quarters of cows with odd numbered ear tags. Cows with even numbered tags served as controls. There were 2182 cases of clinical mastitis for control cows and only 475 for IMD cows. Milk production data analyzed from 1297 completed lactations indicate that cows with IMD produced 1.9 kg more milk per day than controls. The abraded IMD appeared to be well tolerated by dairy cows and abnormal side effects have not been a problem.

Results from these laboratory and field studies on the abraded IMD indicate that the device offers promise in the control of mastitis.

Introduction

Teat dipping and dry cow therapy, two key components of current mastitis control practices, are not effective in the control of intramammary infection by environmental pathogens such as coliform organisms and environmental streptococci. Eighty percent of all intramammary infections caused by coliform organisms result in clinical cases and some of those result in death of the cow (8). The intramammary device (IMD) was developed for the prevention of intramammary infection by environmental mastitis causing pathogens. When inserted into the mammary gland cistern, the IMD stimulates the recruitment of white blood cells (WBC) into the areas of the teat and gland cisterns. This localized recruitment of WBC was shown to provide good protection in prevention of intramammary infection after experimental challenge (23).

A review on the effects of the IMD on quarter milk production and on milk somatic cell counts (MSCC) was presented at the 1984 meeting of the National Mastitis Council (24). The conclusions from that review were that there was a tendency for IMD quarters to produce 0.2 to 0.6 kg less milk per day than control quarters, and that MSCC in IMD quarters were higher than control quarters. The authors concluded that the resulting gain in milk production as a result of reduced mastitis should offset the tendency of IMD fitted quarters to produce less milk. Since that review, modifications to the IMD were made. Laboratory studies and a 6000 cow Israeli field trial were initiated through a USA-Israel Binational Agricultural Research and Development Fund to address questions of the effect of the IMD on milk production, new intramammary infection rate, clinical mastitis, MSCC and tissue damage. This report is intended to summarize, present and interpret those studies.

Laboratory Evaluation

Modifications of the IMD - The original smooth IMD (23) was made of extruded polyethylene, was smooth over its entire surface, and was 2.5 mm in diameter, 115 mm long, and took the form of a loop of 25 mm in diameter in situ. The cannula used for inserting the IMD into the gland cistern was 3.7 mm in diameter and was made of stainless steel. Because of the large diameter of the IMD and cannula difficulty was experienced in inserting the IMD through the teat canal, especially in first calf heifers. Tearing and bleeding of the teat end was commonly observed, even when K-Y jelly was applied to the cannula and teat end. The IMD failed to elevate MSCC in stripping milk to concentrations needed for prevention of infection after experimental challenge (25,26).

The incorporation of finely ground copper into the polyethylene during extrusion of the IMD resulted in an IMD which caused elevations in stripping MSCC to levels considered necessary for protection (17). This smooth copper IMD contained 10% copper, and was 1.5 mm in diameter, 130 mm long and formed a loop of 32 mm in diameter in situ. A stainless steel cannula, 2.4 mm in

diameter, was used to insert the IMD. Reducing the diameter of the IMD and cannula eliminated tearing and bleeding of teat ends. However, copper caused deterioration of the polyethylene and IMD eventually became brittle and broke.

Abrading the surface of the original and molded smooth IMD with medium emery cloth resulted in early prototypes (17) of the abraded IMD (AIMD). Abrading the surface resulted in elevations of MSCC in stripping milk to concentrations that were approximately 60% effective in preventing intramammary infection after experimental challenge (3,19). However, because of the large diameters of the IMD and cannula, cut teat ends, bloody stripping and quarter bucket milk and breakage in situ were observed (3,28).

A smaller diameter polyethylene rod was abraded with medium grade emery cloth to produce the currently used abraded IMD (17). This IMD was 1.5 mm in diameter, 125 mm long, and formed a loop 30 mm in diameter in situ. A stainless steel cannula, 2.4 mm in diameter, was used to insert the IMD. Insertion of the AIMD into the mammary gland cistern resulted in the elevation of MSCC in stripping milk to concentrations that were 68% effective in the prevention of intramammary infection after experimental challenge with Staphylococcus aureus (22). Results (29,30,31) from Israeli field trials reported a 75% decrease in clinical mastitis in cows with AIMD. Discomfort to the cow during insertion has not been a problem. A side effect has been the microscopic appearance of blood in milk from quarters with AIMD (20,22). Bacterial colonization, plaque formation and the appearance of amorphous like material on the surface of the AIMD have been observed (18,20).

To determine if chemical treatment of polyethylene would reduce formation of amorphous like material on the IMD, polyethylene IMD 1.5 mm in diameter and 120 mm long that formed a loop of 25 mm in diameter, in situ, were abraded with medium grade emery cloth or left smooth (20). Abraded and smooth IMD were chemically treated with either carbon tetrafluoride, ammonia, nitric acid, sulfur hexafluoride (SF_6), or left untreated. IMD were individually inserted at random into uninfected mammary quarters and removed 2 months later for observation with a scanning electron microscope. Treatment of IMD with SF_6 appeared to prevent deposits of amorphous like material on the IMD surface without compromising the elevation in stripping MSCC needed for protection (900,000 cells/ml). The abraded SF_6 IMD is currently being tested in the laboratory (9) and in the field (28) to determine effects on bacterial adherence, new infection rate, clinical mastitis, stripping and bucket MSCC and RBC count, and milk production.

Studies are in progress on IMD of differing densities and composition (9,28). Intramammary devices of differing densities are being tested in an attempt to keep the IMD near the floor of the gland cistern, and thus keep the cellular response close to the point of entry of bacteria and further from the mammary secretory tissue.

Scanning Electron Microscope Studies - Seven lactating cows were selected for study (18). The cows had IMD in all 4 mammary quarters. IMD were made of molded polyethylene, 126 mm long and 2 mm in diameter, taking the form of a loop approximately 32 mm in diameter (6).

Foremilk samples were collected aseptically for diagnostic bacteriology and stripping milk for MSCC. Milk samples were collected weekly for 2 wk before IMD removal, at the time of IMD removal and for 3 wk after IMD removal.

Intramammary devices were removed aseptically with alligator forceps from 6 infected and 4 noninfected quarters. The infected quarters were infected with coagulase negative staphylococci, Corynebacterium bovis and a non-specified fungus. The IMD were immediately placed in 2.5% glutaraldehyde in 0.1 M cacodylate buffer, fixed at room temperature for 2 h and at 10 C for 22 h, dehydrated in a graded series of ethanol and critically point dried from CO₂. The IMD were mounted on aluminum stubs and coated with gold palladium in a sputter-coating apparatus. The IMD were kept with dessicant until observed with a scanning electron microscope.

For quarters infected with coagulase negative staphylococci and Corynebacterium bovis and where all of the IMD was retrieved, MSCC decreased from an average of 1.2×10^6 before removal to $.2 \times 10^6$ after IMD removal. In the one quarter where a piece of the IMD remained in the quarter, MSCC remained unchanged at approximately a million cells/ml. For the mammary quarter infected with fungus, MSCC continued to remain in the millions after IMD removal.

The scanning electron microscopy findings indicated that IMD removed from infected mammary quarters contained more amorphous material than did IMD removed from noninfected quarters.

Large concentrations of inflammatory cells were found adhered to abrasions on the IMD surface and to the amorphous material. Deposits of amorphous material were observed forming over inflammatory cells that were adhered to portions of the polyethylene.

Myriads of spherical and rod shaped structures, resembling bacteria, were found within and on the surface of the amorphous material and on the polyethylene surface of IMD removed from mammary quarters infected with coagulase negative staphylococci and C. bovis. Examination of the IMD removed from the quarter infected with fungus revealed branch like structures resembling fungi, growing from deposits on the IMD.

The surfaces of IMD removed from noninfected mammary quarters contained less amorphous material than did IMD from infected quarters. Inflammatory cells were occasionally observed on the surface of the amorphous deposits. The material appeared to be deposited on areas of the IMD that became abraded at the time of insertion into the mammary quarter.

Results from this study suggest that the increased cell counts in infected quarters containing intramammary devices appeared to be associated with microbial colonization the amorphous material.

Milk Somatic Cell Counts - We reported (24) that modest increases of 20,000 cells/ml in quarter bucket milk could be expected during early to mid lactation in cows fitted with AIMD (26). Since then, studies were conducted (16) to determine effects during late lactation. Abraded IMD were inserted into diagonally opposed quarters of 20 cows. Milk somatic cell counts of quarter bucket milk from AIMD quarters were approximately 200,000 cells/ml higher than controls (Table 1). These data suggest that as milk volume in the gland decreases, chemotactic factors generated by the IMD become more heavily concentrated in the milk, causing infiltration of leukocytes further up in the gland and into primary milk.

TABLE 1. Effect of Stage of Lactation on MSCC in Quarter Bucket Milk

Stage of lactation	Quarters		Significance
	Control (Cells $\times 10^3$ /ml)	Abraded IMD	
50 ^a	30	72	P<.05
280 ^b	176	400	P<.05

^a Paape et al., 1984 (19). ^b Paape et al., 1985 (16).

Tissue Irritability - Insertion of either a smooth (20) or AIMD (16) into the mammary gland resulted in a significant increase in the concentration of red blood cells (RBC) in quarter bucket milk (Table 2). The RBC were only visible microscopically. The concentration found in milk from AIMD quarters is equivalent to approximately two drops of blood in a liter of milk. The presence of RBC may be attributable to an abrasive effect of the AIMD to the epithelial lining of the gland cistern. Huston and Heald (10), in studies with the smooth IMD, reported a significant change toward a single layer of epithelial cells in gland cisterns of IMD quarters when compared to controls. Post mortum examination (21) of gland cisterns that had AIMD inserted during lactation, and were microscopically showing RBC in milk, showed pin point hemorrhagic areas in the vicinity where the AIMD was located in situ. Kortum (12) reported more RBC in milk from quarters when the AIMD became lodged in one of the ducts emptying into the gland cistern. A lodged AIMD was assumed on the basis of it not being retrievable with alligator forceps. One can expect this to occur in about 10 to 15% of the quarters that are fitted with IMD (12). The appearance of RBC in milk could also be related to possible disruption of the glandular epithelium caused by increased movement of leukocytes into milk, and to the release of digestive enzymes and toxic oxygen products from phagocytic leukocytes, all of which would contribute towards a more "leaky" glandular epithelium (5).

Table 2. Effect of Smooth and Abraded IMD on Concentration of RBC and NAGase Activity in Quarter Bucket Milk

IMD type	Time relative to insertion	NAGase activity (um/min/ml)	RBC count (X10 ³ /ml)
Smooth ^a	before	.008	26
	after	.011	11,710
	Significance	NS	P<.05
Abraded ^b	before	.014	11
	after	.019	27,335
	Significance	NS	P<.05

^a Mean of 5 mammary quarters. Paape et al., 1984 (27).

^b Mean of 40 mammary quarters. Paape et al., 1985 (23).

The activity of N-acetyl-B-D-glucosaminidase (NAGase), a marker of epithelial cell damage (11), has been measured in quarter bucket milk before and after insertion of IMD (Table 2). There was a tendency for NAGase activity to be higher in IMD quarters after insertion when compared to control quarters. The difference, however, was not statistically significant.

Quarter Milk Yield - Two trials (16) were conducted to determine effects of AIMD on quarter milk yield. In both trials (Tables 3 and 4), there was a tendency for AIMD quarters to produce less milk than control quarters. The difference in milk yield was greater in late lactation (Table 3). The conclusions from that study were that quarter milk yield was too variable a measure for evaluating effects of the IMD on milk production and it is not known to what extent compensatory hypertrophy of control quarters may influence the comparison. Among cow evaluation in field studies involving large numbers of animals, appears to be the more appropriate experimental design to use.

Table 3. Effect of Abraded IMD on Quarter Milk Yield of 9 cows ^{a,b}

Stage of lactation ^c	Quarters		Significance
	Control	AIMD	
	(kg/half/day)		
20 weeks	10.4	10.0	NS
40 weeks	8.4	7.2	NS

^a Mean of 2 milkings. ^b Paape et al., 1985 (23).

^c AIMD inserted 2 weeks postpartum.

Table 4. Effect of Abraded IMD on Quarter Milk Yield of 11 cows ^{a,b}

Stage of lactation ^c	Quarters		Significance
	Control (kg/half/day)	AIMD	
8 month	9.6	9.2	NS
9 month	8.0	7.9	NS

^a Mean of 6 milkings. ^b Paape et al., 1985 (23).

^c AIMD inserted 7 months postpartum.

Immunization and Bacterial Challenge - A study (22) was initiated to determine if immunization would confer any additional protection to mammary quarters with AIMD. In that study AIMD were inserted into diagonally opposed quarters of 14 cows. Seven cows were immunized with a Staphylococcal vaccine. The vaccine was an aqueous solution containing a suspension of formalin killed 2-8 and Smith strains of Staphylococcus aureus, toxin, B toxin, and leukocidin. The vaccine was injected three times, at two week intervals, in the area of the supramammary lymph node. Cows were challenged 2 to 3 weeks later by injecting an AIMD and control quarter with approximately 100 CFU of the 2-8 strain and the remaining two quarters with approximately 50 CFU of the Smith strain. Although an increase in capsular antibody was detected in milk 2 weeks after the second immunization, there were no detectable increases in the ability of neutrophils to phagocytose, in the ability of skimmed milk to support phagocytosis, and in lymphocyte blastogenesis (7,22).

Immunization failed to prevent establishment of infection after intramammary challenge. Among immunized cows, 15 of 23 quarters (65%) became infected and among non-immunized cows, 13 of 22 quarters (59%) became infected. Of 22 AIMD quarters, 7 (32%) became infected, whereas 22 out of 23 (96%) of control quarters became infected. All of the infected quarters were treated with antibiotics (Cefa-Lak) for 14 consecutive milkings. In the next lactation, 5 out of 7 previously infected AIMD quarters were still infected, whereas none of the 22 previously infected control quarters were infected (4). These preliminary data suggest that where the AIMD has failed to protect against a staphylococcal infection, its continued presence may render therapy more difficult. Although the effects of immunization on enhancement of immunological function and prevention of intramammary infection were disappointing, a recent study (27) using an oil suspension of this vaccine obtained a 90% protection rate after experimental challenge.

Field Evaluation

The AIMD is being tested (29,30,31) in a completely randomized-split herd study in 13 commercial dairy herds in Israel. Herds selected are typical for the national dairy herd. All are free from Streptococcus agalactiae and the levels of infections due to other streptococci and S. aureus range between 5 and 12% and 8

and 26%, respectively. In September 1986, this field trial will be entering its fourth and final year.

Milk somatic cell counts, milk production, new intramammary infection rate and incidence of clinical mastitis will be determined on 3000 cows with AIMD in all 4 quarters and on 3000 control cows. Intramammary devices measuring 1.5 mm in diameter, 125 mm long, forming a loop 30 mm in diameter in situ, and abraded with medium grade emery cloth are being used. The AIMD are inserted with a stainless steel cannula, 2.4 mm in diameter, into all 4 quarters of cows with odd numbered ear tags. Within a herd, all of the AIMD are inserted on the same day regardless of stage of lactation or dry period. Cows with even numbered ear tags serve as controls. Cows are observed at each milking by either the herdsman or milker for signs of clinical mastitis. Clinical cases with systemic involvement are diagnosed by the veterinarian.

Quarter foremilk samples are taken before and every three months after AIMD insertion for diagnostic bacteriology. Weigh jar, stripping and bulk tank milk samples are taken before and every three months after AIMD insertion for MSCC.

Milk Somatic Cell Counts - The effect of the AIMD on MSCC in 72 noninfected primiparous cows was monitored monthly throughout lactation (Table 5). Mean MSCC in foremilk and stripping milk from AIMD quarters were greater than one million/ml during the entire 9 months of observation. Weigh jar MSCC were never greater than 500,000/ml. Foremilk and stripping MSCC from AIMD were higher ($P < .01$) than controls. There was no difference in weigh jar MSCC between AIMD and control cows. Milk somatic cell counts of greater than one million were found among 95.3% of the stripping milk samples from AIMD quarters (Table 5). The proportion of quarters having MSCC of greater than one million in stripping milk gradually decreased to 36.6% by 9 months. These data indicate that the AIMD was capable of eliciting and maintaining potentially protective levels of leukocytes in stripping milk from at least 70% of the quarters for at least 6 months without causing an unacceptable rise in bucket MSCC.

Tissue Irritability - The AIMD appeared to be well tolerated and safe in non-infected quarters of lactating cows (39,41). A low yet transient incidence of clots and flakes in foremilk and strippings was observed after AIMD were inserted into subclinically infected quarters. The intensification of the MSCC response to the AIMD in subclinically infected quarters supports similar findings from an earlier field study (7) with the smooth IMD. In the Israeli study, even though subclinically infected cows received AIMD, no increase in the bulk tank MSCC among the herds on the study was observed (39). Bulk tank MSCC counts in those herds averaged between 150,000 to 200,000/ml. Evidently, the higher cell counts in milk from subclinically infected AIMD quarters was obscured due to dilution with milk from noninfected AIMD and control quarters.

Slightly bloody secretions were found (41) in samples removed 2-8 weeks after drying off in 6 quarters (1.5%) of 98 AIMD cows and

in 4 quarters (1.4%) of 73 control cows examined. Slightly pink-tainted colostrum was observed in 11 quarters (1.5%) of the 186 cows fitted with AIMD at drying off but also in 6 quarters (0.8%) of the 194 control cows.

Table 5. Effect of Abraded IMD on MSCC in Milk Fractions From 72 Noninfected Primiparous Cows^a

Time Relative to AIMD	Milk Fraction	Percent of the quarters with MSCC > 10 ⁶ /ml					
		AIMD	Control	AIMD		Control	
				Mean	SE	Mean	SE
(x 10 ³ /ml)							
Before (72 cows)	fore	0	0	180	38	228	41
	strippings	0	0	75	72	311	65
	weigh jar	0	0	92	16	76	20
After 2 wk (72 cows)	fore	88.6	0	3774	181	217	19
	strippings	95.3	0	5960	265	386	45
	weigh jar	0	0	390	48	108	12
4 wk (68 cows) ^b	fore	68.0	0	2967	138	240	25
	strippings	83.3	0	4980	318	385	62
	weigh jar	0	0	388	65	260	27
3 mo (63 cows)	fore	54.2	0	1905	250	318	36
	strippings	79.4	0	2876	311	412	51
	weigh jar	0	0	406	72	318	46
6 mo (52 cows)	fore	42.1	0	1625	278	420	47
	strippings	71.8	0	2150	196	617	38
	weigh jar	0	0	480	52	327	50
9 mo (49 cows)	fore	23.6	8.6	1077	311	890	97
	strippings	36.6	17.2	1566	289	1072	160
	weigh jar	0	0	511	120	473	88

^a Ziv et al., 1984 (30).

^b Number of cows progressively reduced due to culling or drying.

Milk Yield - Milk production data was recently analyzed (14) from 1297 lactations from Israeli cows in 11 herds which completed one lactation since AIMD were inserted. Cows with AIMD produced 148 kg more milk in a total lactation than did control cows (Table 6). Milk production per day was even more favorable for the advantage of AIMD cows (1.9 kg/day). These data agree with previous results (13) that was based on monthly test day information from three of these herds. The larger difference in milk per day comes about because AIMD cows had shorter lactations by about 18 days. Reasons for the shorter days in milk for AIMD cows will not be resolved until information relative to clinical mastitis, reproduction and culling among the cows within these herds are analyzed. It was suggested (12) that an increased incidence of acute clinical coliform mastitis during early lactation in control cows could have delayed breeding. Also, the reported (16) increase in MSCC in AIMD quarters during late lactation may have contributed to an earlier involution of the mammary secretory tissue.

Table 6. Effect of Abraded IMD on Lactation Milk Yield^{a,b}

Measure	Controls	AIMD	Significance
Lactation milk (kg)	7889	8037	P<.05
Milk/day (kg)	23.7	25.6	P<.01
Days in milk	336	318	P<.01

^a Miller et al., 1986 (14).

^b Data obtained from 1,297 lactations.

Table 7. Milk Yield for 11 of 13 Herds Used in the Israeli Abraded IMD Field Study

Herd	Treatment	Milk Yield (kg)	Standard Error
1	Control	9000	304
	AIMD	9374	292
2	Control	9552	200
	AIMD	9417	210
3	Control	7278	335
	AIMD	7304	390
4	Control	6859	333
	AIMD	7344	334
5	Control	7274	304
	AIMD	7513	304
6	Control	6816	288
	AIMD	7046	253
7	Control	7266	277
	AIMD	7409	241
8	Control	8132	168
	AIMD	8323	173
9	Control	7564	146
	AIMD	7531	157
10	Control	8126	191
	AIMD	8428	183
11	Control	8103	154
	AIMD	8498	156

^a Miller et al., (14).

Data thus far indicate that one can expect higher milk production for cows with AIMD in the first lactation after insertion. The increase appears to be based on the reduction in clinical mastitis in AIMD cows. However, it appeared that there are some herds where the benefit to milk production was not present (Table 7). It is possible that use of the AIMD may be of no benefit in well-managed herds with initial low infection rates.

New Intramammary Infection Rate - Effects of the AIMD on new intramammary infection (IMI) are shown in Table 8. New IMI due to Non-agalactiae streptococci and Corynebacterium bovis were lower for AIMD cows when compared to controls (29). New IMI due to E. coli infections were reported (30) to be lower in AIMD

cows. Infections due to coagulase negative staphylococci were higher in AIMD cows (Table 8). There was no difference between AIMD and control cows in new IMI caused by Staphylococcus aureus. More recent data (29) obtained from 4897 cows, that included 3152 with 2 full lactations (Table 12), indicated no difference between AIMD and control cows in new IMI. However, the spontaneous cure rate for all existing IMI was significantly higher ($P < .05$) in AIMD cows (29). For AIMD cows, the cure rate averaged 1.25/100 cows/month and for controls 0.25/100 cows/month. These data suggest that the advantage gained in reduced new IMI among AIMD cows was lost in subsequent lactations but may be offset by the increase in spontaneous cures among AIMD quarters. Although data are still limited, a trend towards fewer new IMI acquired during the dry period was reported (30) among AIMD cows when compared to controls (Table 9).

Table 8 New Intramammary Infections (IMI)^a

Organism	New quarter IMI/100 Cows/month ^b		
	AIMD Cows (%)	Control Cows (%)	Significance
<u>S. aureus</u>	2.7	2.3	NS
Coag. neg. staph.	3.6	2.9	$P < .06$
Non-agal. strep.	1.5	2.1	$P < .05$
<u>C. bovis</u>	0.9	1.4	$P < .05$
Others	1.7	1.9	NS
Total	10.4	10.6	NS

^a Ziv, 1985 (29).

^b Data obtained from 3,660 cows.

Table 9. New Intramammary infections (IMI) acquired during the dry period after Abraded IMD were placed at time of off time^a

Herd		No. of cows	Percent new IMI due to			Total
			Non-agal. strep	<u>S. aureus</u>	Others	
7	IMD	95	1.58	2.63	0.53	2.11 ^b
	Cont	91	3.03	3.57	0.27	6.87
8	IMD	91	1.10	4.95	0.81	6.86 ^c
	Cont	103	1.70	6.31	1.21	9.22

^a Ziv et al., 1985 (30).

^b Significantly less than control ($P < .01$).

^c Significantly less than control ($P < .05$).

Clinical Mastitis - Sixty-nine percent of all clinical cases of mastitis (530 cases) occurred in control cows (Table 10). The most dramatic difference occurred for systemic clinical cases that required antibiotic therapy, where control cows accounted for 83% of all reported cases. The difference between AIMD and

control cows was not as dramatic for clinical cases not requiring antibiotic therapy. Fifty-eight percent of those cases occurred in control cows. It is possible that a number of those clinical cases occurred during the few weeks after inserting AIMD in infected cows.

Inserting an AIMD into infected mammary quarters will result in intensification of the MSCC (6), resulting in the transitory appearance of garget in foremilk from some of those quarters (30).

Table 10. Frequency of Clinical Mastitis in Israeli Abraded IMD Field Study^a

<u>Clinical Cases</u>	<u>AIMD Cows^b</u>	<u>Control Cows^c</u>
Systemic Involvement	41	197
No Systemic Involvement	123	169
Total	164	366

^a Ziv et al., 1985 (42).

^b Data obtained from 1,998 cows.

^c Data obtained from 2,002 cows.

Bacteriological examination of aseptically collected foremilk samples from AIMD and control cows with systemic clinical mastitis indicated that 70% of the isolates were identified as coliforms (31). From clinical cases with no systemic involvement, 70% were identified as S. aureus.

The frequency of clinical mastitis with systemic involvement in AIMD cows relative to the last stripping MSCC is shown in Table 11. Ninety-five percent of the cases occurred when the MSCC in stripping milk was below 500,000/ml. None occurred when the cell count was over 750,000/ml. These data indicate that for maximum protection, the AIMD must elevate the cell count in stripping milk to at least 750,000/ml. This is very close to the level (900,000/ml) reported (26) necessary for protection in laboratory studies. The data also indicate that the AIMD failed to elevate stripping MSCC to protective levels in some cows. In studies (15) at Beltsville, 56% of AIMD quarters had MSCC greater than 500,000/ml and 33% were below 200,000/ml (non-responders). It would appear that even greater reductions in clinical mastitis could be achieved by reducing the number of non-responders within a herd.

As of March 31, 1985, 4897 cows were enrolled in the AIMD Israeli field study (Table 12), with 3152 cows completing two full lactations (40).

Table 11. Frequency of Clinical Mastitis with Systemic Involvement in IMD Cows Relative to Last Stripping MSCC^{a,b}

MSCC x10 ³ /ml	Cases	
	No.	(%)
<200	24	58.5
201-500	15	36.6
501-750	2	4.9
>750	0	0.0
Total	41	100.0

^a 1-89 days before onset of a case.

^b Ziv, 1985 (29).

Table 12. Cows on Test in Israeli Abraded IMD Field Study as of March 31, 1985^a

Group	Length of Lactation	No. of Cows
1	Completed 2 full lact.	1284
2	Completed 2 full lact.	1868
3	Observed for 7 months	915
4	Observed for 4 months	830
Total		4897

^a Ziv, 1985 (29).

Of the 2657 reported cases of acute and mild clinical mastitis, only 18% or 475 cases occurred in AIMD cows (Table 13). Of the 475 cases in AIMD cows, 91% occurred in cows that completed 2 full lactations, suggesting that the AIMD may become less effective in later lactations. Stripping MSCC in AIMD quarters reported in Table 5 showed that the percentage of quarters with stripping MSCC of greater than one million/ml, decreased from a high of 95.3% 2 weeks after insertion to 36.6% 9 months after insertion. This suggests that with advancing lactation, the AIMD loses its ability to maintain MSCC in stripping milk at levels needed to prevent clinical mastitis.

Table 13. Total Cases of Acute and Mild Clinical Mastitis^a

Group	AIMD Cows		Control Cows	
	No.	%	No.	%
1	245	21.0	1090	79.0
2	186	22.4	635	77.6
3	28	8.0	316	92.0
4	16	10.2	141	89.8
Total	475	17.8	2182	82.2

^a Ziv, 1985 (29).

The reduction in the prophylactic effect of the AIMD with advancing lactation is supported by data presented in Table 14. Among AIMD quarters twice as many acute clinical cases occurred during the 12 to 24 month period after insertion when compared to the first 12 months after insertion.

Table 14. Acute Clinical Mastitis According to Time Since Abraded IMD Insertion (March 31, 1985 Update)^a

Months after IMD Insertion	Cases/100 cows/month		Ratio Control:AIMD
	AIMD	Control	
<3	1.8	9.3	5.2
3-6	1.2	7.2	6.0
6-9	2.1	11.6	5.5
9-12	1.3	8.3	6.4
12-15	3.8	13.1	3.4
15-18	4.6	9.2	2.0
18-21	4.3	12.7	2.9
21-24	4.8	10.0	2.4

^a Ziv, 1985 (29).

Summary

Earlier versions of the smooth and abraded IMD either failed to prevent establishment of infection or produced side effects such as the appearance of blood in milk. The currently used abraded IMD is 1.5 mm in diameter, 125 mm long, and forms a loop 30 mm in diameter in situ. This AIMD has been tested experimentally and was shown to provide good protection against establishment of infection after experimental challenge.

The AIMD is currently being field tested in Israel in a study involving approximately 5000 cows. To date, significant reductions in clinical mastitis have been observed, especially acute clinical coliform mastitis. Further, AIMD cows are producing an average of 1.9 kg more milk per day than control cows. The AIMD appears to be well tolerated by dairy cows and abnormal side effects have not been a problem. Veterinarians and dairymen are convinced over the benefits of the AIMD, and dairymen are willing to pay to have cows fitted with AIMD.

While results from experimental and field investigations appear promising, the authors feel that a thorough and complete analysis of the results from this BARD study and from BARD No. US-854-84, a one year study on the IMD, must be made before practical recommendations for use in the United States can be made.

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Description of Cooperation

Excellent cooperation was in evidence between laboratories in Israel and USA. Joint papers were prepared and presented by Dr. Ziv at XIII World Congress on Diseases of Cattle in Durban, South Africa, 1984; by Dr Ziv at Seminar on "Progress in the Control of Bovine Mastitis" in Kiel, Germany, 1985; by Dr. Paape at 23rd Annual Meeting of the National Mastitis Council, Inc. in Kansas City, Missouri, 1984; and by Dr. Paape at 25th Annual Meeting of the National Mastitis Council, Inc., Columbus, Ohio, 1986.

In 1983, Dr. Paape spent one month in Israel and in 1984, Dr. Ziv spent one month at Beltsville where results from the field trial in Israel and laboratory studies at Beltsville were thoroughly reviewed. In addition Drs. Paape and Ziv spent one week together in Kiel, Germany going over results from IMD studies. As a result of Dr Ziv's international recognition, Dr Ziv was in the USA several times during the BARD agreement, and always spent a few days to a week at Beltsville reviewing results and presenting seminars on the IMD Israeli field trial.

Because of this excellent cooperation, the project was very successful, is recognized by the international scientific community as innovative research that resulted in important basic findings which led to practical application in the control of mastitis.

Benefit to Agriculture

The dairy cow has evolved into the major supplier of milk as a human food source. In the United States alone there are approximately 215,000 dairy farmers and around 11,000,000 cows, while on a worldwide basis the total number of cows in selected western and eastern countries approximates 102,000,000 cows.

Mastitis is a chronic problem on dairies of all sizes, in all areas. In the average herd 50% of the cows are infected in half of their quarters. The effect on an infected animal can range from a mild reduction in the quantity and quality of milk produced to the death of the cow. Typically, the result is an economically significant reduction in milk quantity and quality. Treatment with antibiotics is then needed to restore quality and productivity, and to prevent the spread of infection to the rest of the herd. The National Mastitis Council in 1983 estimated that mastitis costs dairymen \$200/cow/year. In the United States this would represent a yearly loss of over two billion dollars.

All recent data indicates that the IMD can reduce mastitis by at least 60%. Hence, savings of at least \$120/cow/year can be realized. Use of the IMD offers the dairy industry the possibility of dramatic increases in productivity and efficiency heretofore not available. Also, its usage holds the additional prospect of controlling mastitis caused by environmental pathogens in a manner that is economically an improvement on today's methods.

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