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A randomized controlled trial to compare the use of homeopathy and internal Teat Sealers for the prevention of mastitis in organically farmed dairy cows during the dry period and 100 days post-calving

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Introduction: Routine use of antibiotics to prevent mastitis in dairy cows is prohibited by organic farming regulations. Internal Teat Sealers have been proposed as an alternative. We compared two drying-off (DO) supporting measures (Internal Teat Sealer and Homeopathy) to an untreated control group to assess their protective effects against clinical mastitis and intra-mammary infections during dry period of dairy cows.

Methods: A field trial with 102 dairy cows from 13 Swiss organic dairy farms was conducted. Cows were randomly assigned to one of three groups within a herd. In the Internal Teat Sealer group (ITS; 36 cows) cows were treated with the commercial ORBE-SEAL (Pfizer) in all four quarters immediately after the last milking. In the Homeopathy group (HDT; 32 cows) the cows were treated per-orally by a herd-specific homeopathic formulation consisting of two remedies in 1:10⁶ dilution over 5 days before and after DO. The untreated group received no therapy (U; 34 cows).

Results: For ITS, HDT and U the clinical mastitis incidence rates for the first 100 days post-calving were 11%, 9% and 3%, respectively, and the proportion of normally secreting quarters was (quarter somatic cell count (SCC) [QSCC] < 100,000/ml) 70%, 68%, and 65%, respectively. Power analysis indicates that a proportion of 75% would support the rejection of null hypothesis in the HDT, and 74% in the ITS group against untreated control. Quarters of cows with SCC < 200,000/ml at DO showed significantly higher normal secretion in HDT group (odds ratio [OR] 9.69) compared to untreated control, whereas Teat Sealing lead to an OR of 3.09 (not significant, *post hoc* power 31.3%).

Conclusions: Under the studied conditions herd-specific homeopathic dry cow therapy was effective in increasing the number of animals with normal milk secretion after subsequent parturition, compared to untreated control. It may be an effective alternative to Teat Sealing, particularly in animals with relatively low SCCs. Further research is required to confirm these results, and under different environmental conditions. *Homeopathy* (2010) 99, 90–98.

Keywords: Dry cow therapy; Dairy cow; Mastitis; Teat Sealer; Homeopathy; Organic farming

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Introduction

The organic farming regulations in the EU (2000) and in Switzerland (2001) provide the framework for organic herd health management: farmers and veterinarians are encouraged to reduce chemical drug usage and residue accumulation in the environment.^{1,2} Nevertheless, the generally preferred approach for maintaining udder health is therapeutic and preventive application of antibiotics,³ known as 'dry cow therapy' (DCT). DCT applied at the end of lactation is expected to provide two benefits. Firstly, the period of treating sub-clinically infected cows with antibiotics is shifted to a period without milk production, reducing the economic loss from discarded milk.⁴ The second benefit of antibiotic DCT is prevention of proliferation of invading bacteria after drying-off (DO), especially during the first 2 weeks when the infection risk is very high.

The search for alternatives to antibiotics is driven by increasing consumer demand to avoid chemical drugs, reducing the potential risk of residues in the food chain⁵ and possible promotion of resistant pathogens such as multi-resistant *Staphylococcus aureus*.⁶ Teat Sealing is an antibiotic reduction measure invented nearly 30 years ago.⁷ The first products were latex based sprays which hardened after external application to the teat end to prevent invasion by environmental pathogens (*Streptococcus uberis*, *Coliforms* and *Enterococci* etc.). Initial results indicated these so called External Teat Sealer (ETS) could achieve significant reduction of intra-mammary infections (IMIs) for certain microorganisms.⁷ Recent investigations demonstrated adherence problems which reduce the effect duration to only 3 days.⁸ A lactation-terminating iodine dip is cheaper and equally effective.⁹

Internal Teat Sealants (ITSs) consisting of bismuth subnitrate, a viscous temperature stable substance, is simple to apply. The substance is assumed to seal the teat against microbial invasion by providing a physical barrier during the entire dry period.¹⁰ ITSs are recommended as an antibiotic-free udder health strategy for organic dairy systems.^{5,11} Early investigations stressed a significant risk reduction of IMIs during dry period by ITS compared to untreated udder quarters and nearly equal protective effects compared to antibiotic DCT,^{12,13} particularly against environment associated pathogens like *S. uberis*.¹⁴ The manufacturer's recommendation is to treat only cows with a low risk of sub-clinical infection (low somatic cell count [SCC]). Combined therapy using antibiotics combined with ITS is the most recent research field¹⁵⁻¹⁷ but does not match the organic requirements for reduced antibiotic use and can be costly.

Homeopathy is one of the preferred methods treating mastitis in organic farming.¹⁸ Though there is no evidence for efficacy of potentized substances in bovine mastitis, farmers rely on these because: they are easily administered, inexpensive, often recommended by friends and family and do not require withholding of milk after treatment.¹⁹ While some studies concerning clinical mastitis treatment by homeopathy have been published, the prophylactic use is limited to the application of so called nosodes, which

have not been shown to be effective in cure or prevention of sub-clinical mastitis during lactation.^{20,21} In a German study homeopathic combinations given in addition to antibiotics at DO, reduced the rate of subsequently infected quarters significantly. Treating cows additionally to antibiotics, post-partum infection rates of only 17% were achieved compared to 70% in the placebo control group, although there was no difference in infection rates in healthy quarters, compared to placebo treatment without antibiotics.²² The lack of studies about homeopathy at DO complicates the search for candidates for homeopathic treatment of cows at this time. In contrast to finding an individual remedy, the concept of herd-specific treatment considers the farm, herd and predominant animal profile. This approach is part of the DO strategy in the Swiss Organic Dairy Farmer's Network "pro-Q".

This investigation evaluates three treatment approaches (homeopathic DCT, Internal Teat Sealing versus an untreated control group) in a prospective randomized comparative study. The hypothesis was that homeopathy is an effective method to reduce dry period IMI compared to therapy omission and achieves comparable results to ITS. The trial was conducted in order to test the hypothesis that homeopathy is able to reduce the incidence of sub-clinical mastitis during the peri-parturient period as indicated by the isolation of mastitis pathogens from milk samples, or the elevation of SCCs. A secondary outcome looked at the effects of homeopathic treatment on the incidence of clinical mastitis during the dry period and 100 days post-calving.

Material and methods

In 13 Swiss organic dairy herds a total of 102 cows at DO were enrolled into the study. Twice in the last 10 days before DO, sterile milk samples were taken from all four-udder quarters. After bacteriological and clinical examination of the milk samples, cows were randomly allocated to the three different DO treatment groups using a randomized block design at herd level (3-6 cows per-herd represented one block). Randomization was conducted by using single-digit random numbers (1-9 omitting zero) generated by Microsoft Excel[®]. Any cow with a chronic infection involving major udder pathogens, diagnosed by previous culture during lactation, was excluded.

Sub-clinical mastitis in at least one of the samples was defined as 'sub-clinical'. In case of differing microbiologic diagnosis between the two samples, a third milk sample was taken. In case of negative findings in both milk samples the SCC of the second sample determined the diagnosis 'normal secretion' (SCC < 100,000/ml) or 'increased SCC' (SCC ≥ 100,000/ml), respectively.

All cows were dried-off abruptly and treated as follows:

Group ITS: Teat Sealer Orbeseal (Pfizer Inc., Zurich) applied to all four quarters ($n = 36$ cows with 144 quarters). After cleaning and disinfection of the teat immediately after the last milking, the Orbeseal application device was partially inserted into the teat canal and 4 g of Bismuth subnitrate administered into the teat.

Group HDT: The homeopathic DCT group received a homeopathic medicine standardized at herd level ($n = 32$ with 128 quarters). The homeopathic herd remedy was determined by consideration of the predominant cow type according to homeopathic Veterinary Materia Medica (Steingassner,²³ Wilhelm Maudrich Verlag, Vienna, Munich, Berne, 1998). This was determined using a herd profile questionnaire covering the main factors describing physical cow type (height, body habitus, tissue character etc.), predominant herd diseases (fertility disorders, mastitis, extremity problems or metabolic disorders) and behaviour and temperament of the herd (see Table 1). The best fitting remedy represents the herd-specific homeopathic. Eight remedies were used for the 13 herds: *Mercurius solubilis*, *Lachesis mutus*, *Sulfur*, *Calcium carbonicum*, *Calcium phosphoricum*, *Pulsatilla pratensis*, *Sepia*, *Silica*, all 6x.

Once a day over the last 5 days of lactation cows were treated orally with 10 lactose globuli impregnated with the herd remedy and additionally 10 globuli containing the homeopathic *Silica 6x* (WELEDA, Arlesheim) dispersed in tap water in a 10 ml syringe. The syringe was agitated to distribute the globuli equally. One week later the treatment cycle was repeated.

Group U: Untreated control group ($n = 34$ with 136 quarters). This group was dried-off abruptly with no interventions other than post-milking teat dipping, as conducted routinely after each milking.

In order to investigate the intervention prognosis dependent on udder health at DO, the study population was also sub-classified at a quarter level, on the basis of milk samples prior to DO and the last routine milk recording, respectively:

- bacteriologically negative (BAC...)
- <100,000 cells/ml (QSCC100...)
- <200,000 cells/ml (QSCC200...)
- <200,000 cells/ml in the last routine milk record (CSCC200-).

The latter criterion was included since the commercial recommendation for Teat Sealing is that SCC should not exceed 200,000/ml, in order to minimize the risk of clinical dry cow mastitis.

Seven to fourteen days after calving, quarter milk samples from each trial cow were taken for bacteriological and cell count investigation. The quarter diagnosis was classified to categories: (1) normal secretion (NORM), (2) increased SCC with >100,000 cells/ml (SCCI), (3) infected by minor pathogens (MIPI; *Staphylococcus* spp.), (4) infected by cow associated (not environment associated) major pathogens (MAPI; *S. aureus*, *S. agalactiae* and *S. dysgalactiae*, *Streptococcus* spp.), and (5) infected by environment associated major pathogens (EAPI; *S. uberis*, *Enterococci*, *Coliform* bacteria). For these five classifications the distribution in each treatment group was calculated for all treated quarters and the sub-categories described above, separately.

Cow SCC (CSCC) comparison using monthly routine milk records was performed after logarithmic transformation of values to Linear Somatic Cell Score (LS) using formula (1):

$$LS = \log_2(\text{SCC}/100,000) + 3 \quad (1)$$

Statistical analysis

To compare **TREATMENT** effects the Pearson χ^2 was calculated. For quantification of the treatment effects influenced by udder health factors prior to DO, a logistic regression model was fitted to the binary dependent variable **NORMAL** (for normal secretion after calving: SCC in the milk sample <100,000/ml and no pathogens) and the independent variables **TREATMENT** and **BAC** (bacteriological findings at DO with the levels (1) no cultural findings, (2) minor pathogens and (3) major pathogens, definitions above), **QSCC** (quarter SCC of the milk sample prior with levels (1) <100,000/ml, (2) 101–200,000/ml and (3) >200,000/ml at DO), and **CSCC** (cow SCC with levels (1) <200,000/ml in last milk recording date before DO and (2) $\geq 200,000$ /ml) including interaction terms.

The variable levels with highest hypothetical negative impact on the dependent variable served as reference terms ('untreated' for **TREATMENT**, 'SCC $\geq 200,000$ /ml' for **QSCC** and **CSCC**, 'major pathogens' for **BAC**). To calculate the best fitting model, the Akaike Information Criterion was used after stepwise reduction of the model by omitting non-significant variables including their interaction terms. The statistical report on the final models included z Statistic, odds ratio (OR) in comparison to the reference level (to be interpreted as 'chance of protection') and 95% confidence intervals.

Statistical significance of differences between mean least square of the respective sample month (3 months prior to DO and 3 months after calving) was calculated by ANOVA.

For statistical analysis the general significance level was set to $\alpha = 0.05$. All data analyzed by the statistic software package CRAN R ver 2.9 (R Development Core Team, 2009). *Post hoc* power analysis was performed using G*Power software to determine actual statistical power in case of marginally non-significant results.

Results

No clinical mastitis cases of mastitis were observed in the dry period. During 100 days of subsequent lactation, a total of eight clinical mastitis cases were observed. No significant differences among treatment groups were found, but the untreated control showed the lowest incidence of mastitis (3% vs. 9% and 11%, for U, HDT and ITS, respectively). Due to the study design (to include sub-clinically infected quarters before treatment), a certain number of bacterial infections were found in the last investigation before DO the cows. The culture results are shown in Table 2.

Nearly 80% of all quarters were cultured negative ($n = 324$ of 408) ranging from 76 to 83% in the three treatment groups, with no significant difference between groups. The most common pathogens found in the samples were coagulase-negative *Staphylococcus* spp. (8–13%, mean 11%) followed by *S. aureus* (1.5–5%; mean 3.7%) and *S. uberis* (2–4%; mean 3.4%).

Table 1 Questionnaire and repertory for herd-specific remedy determination. Each x represents one point per remedy in case of frequent appearance within the herd according to farmer's assessment; highest sum of points indicates the herd remedy

Cluster	Area	Herd symptom	Remedy															
			Calc	Phos	Puls	Nux-v	Sep	Sulphur	Calc-p	Lach	Lyc	Merc	Nat-m	Sil				
Appearance	Body	Bulky	X					X										
		Thin		X			X		X			X						
		Stumpy		X			X		X			X						
		Big		X			X		X			X						
		Meagre				X	X					X	X	X	X			
	Head	Small/fine			X						X							
		Big	X							X		X					X	
		Small			X							X						
	Belly	Normal		X		X	X	X	X			X		X	X			
		Big	X					X				X					X	
	Udder	Atonic							X									X
		Not atonic	X	X	X	X			X	X	X	X	X	X	X	X		
	Coat	Dirty							X	X	X			X			X	X
		Not shiny							X	X						X		
		Shiny		X														
		Very bright	X		X								X	X				
	Herd age	Normal				X						X	X					
		Many young cows				X						X	X					X
Many old cows						X					X	X					X	
Behaviour	Character	Average	X	X		X		X	X	X			X	X	X			
		Dominant	X	X			X					X	X					
		Irritable				X		X			X	X			X			
		Curious		X						X								
		Nervous		X		X				X			X				X	
		Agile		X		X				X	X		X					
		Sensitive		X		X		X	X		X	X					X	
		Anxious		X						X	X	X					X	
		Shy	X		X										X		X	
		Trusting			X					X								
		Gentle			X										X			
		Calm	X		X				X		X				X			
	General modalities	Stubborn	X				X	X				X			X			X
		Greedy	X			X						X			X			
		Unconcerned					X						X					
		Calf separation			X										X			
		Cold aggravates	X				X			X				X				X
		Heat aggravates			X				X			X				X		
Diseases	General	Infections	X	X	X				X		X							
		Suppuration	X								X		X					X
	Milk	SCC high	X	X					X				X					X
		Incontinentia lactis	X											X				
		Milk withdrawal		X		X	X					X						
	Skin/Tissue	Skin problems			X		X	X					X	X				X
		Warts	X													X		
		Weakness of ligaments					X							X				X
		Joints	X							X				X	X			
		Claws							X						X			X
	Genitalia	Vaginal prolapse					X											X
		Fertility problems		X	X		X					X			X			
		Metritis			X		X					X						
	Obstetrics	Dystocia			X		X											
		Retentio secundinarum					X					X						
	Metabolism	Ketosis		X		X							X					
		Hypocalcemia	X			X					X	X						
		Diarrhia	X	X					X	X								
Ruminal tympany					X				X			X	X					
Respiratory	Diseases	X	X	X	X			X			X	X						

Table 2 Bacteriological quarter profile within treatment classes before DO and after calving (CLV); no. of quarters (%); differences between treatment groups and sample date not significant

Bacteriological diagnosis		Treatment group						Total	
		No. of cows (%)							
		HDT		ITS		U			
		DO	CLV	DO	CLV	DO	CLV	DO	CLV
Negative		97 (75.8)	104 (81.3)	119 (82.6)	118 (82.0)	108 (79.4)	110 (70.9)	324 (79.4)	332 (81.4)
Minor pathogens (MIPI)	<i>Staphylococcus</i> ssp.	17 (13.3)	2 (1.6)	12 (8.3)	10 (6.9)	16 (11.7)	10 (7.4)	45 (11.0)	22 (5.4)
Major pathogens (MAPI)	<i>Staphylococcus aureus</i>	6 (4.7)	8 (6.3)	7 (4.9)	7 (4.9)	2 (1.5)	1 (0.8)	15 (3.7)	16 (3.9)
	<i>Streptococcus</i> ssp.	1 (0.8)	–	1 (0.7)	2 (1.4)	– (0.00)	1 (0.7)	2 (0.4)	3 (0.7)
Environment associated pathogens	<i>Streptococcus uberis</i>	5 (3.9)	11 (8.6)	3 (2.1)	6 (4.2)	6 (2.9)	6 (4.4)	14 (3.4)	23 (5.6)
	<i>Enterococci</i>	–	–	–	–	–	4 (2.9)	–	4 (1.0)
	<i>Coliforms</i>	–	2 (1.6)	–	1 (0.7)	–	3 (2.2)	–	6 (1.5)
	<i>Proteus</i>	–	–	–	–	–	1 (0.7)	–	1 (0.25)
Others		2 (1.6)	1 (0.8)	2 (1.4)	–	4 (2.9)	–	8 (2.0)	1 (0.25)
Total		128	128	144	144	136	136	408	408

After calving the profile changed slightly (Table 3). No significant differences were found in the number of negative quarters (79.4% at DO and 81.4% after calving) with similar numbers in the three treatment groups. The rate of *S. aureus* infections was nearly the same as the pre-drying profile (3.7% at DO and 3.9 after calving). Numbers of infections caused by environment associated pathogens (EAPI) increased during the dry time. *S. uberis* was found in 23 of samples after calving (compared to 14 before DO) and *Enterococci* not diagnosed before DO was found in four quarter milk samples of the untreated control. Additionally, an analysis of quarters with formerly negative bacterial findings was performed ($n = 324$). The results are tabulated in Table 3.

Table 3 shows that the trend of more frequent *S. uberis* infections in group HDT compared to the entire population,

indicating limited protective effects of homeopathic therapy against this pathogen, in contrast to the development of *Staphylococcus* ssp. For all *streptococci* and *staphylococci*, the untreated control group showed the lowest new infection rates. In order to understand the protective effects of the chosen treatment strategies, diagnoses of post-partum samples were sub-categorized depending on cell count and pathogen class at DO. The results according to different restriction criteria (all, uninfected quarters, quarters below a certain SCC threshold and quarters of cows with low SCC) are shown in Table 4.

Considering all involved quarters the proportion of normally secreting quarters is 68% (87 out of 128), 70% (101 out of 144), and 65% (110 out of 136) for group HDT, ITS and U, respectively. There is no statistically significant difference between groups, power calculation

Table 3 Bacteriological quarter profile within treatment classes after calving in previously negative quarters

Bacteriological diagnosis		Treatment group			Total
		No. of cows (%)			
		HDT	ITS	U	
Negative		80 (82.5)	98 (82.3)	92 (85.2)	270 (83.3)
Minor pathogens (MIPI)	<i>Staphylococcus</i> ssp.	2 (2.1)	8 (6.7)	7 (6.5)	17 (5.3)
Major pathogens (MAPI)	<i>Staphylococcus aureus</i>	3 (3.1)	5 (3.4)	1 (0.9)	8 (2.5)
	<i>Streptococcus</i> ssp.	0	2 (1.7)	1 (0.9)	3 (0.9)
Environment associated pathogens (EAPI)	<i>Streptococcus uberis</i>	10 (10.3)	6 (5.0)	2 (1.9)	18 (5.6)
	<i>Enterococci</i>	0	0	2 (1.9)	2 (0.6)
	<i>Coliforms</i>	1 (1.0)	1 (0.8)	2 (1.9)	4 (1.2)
	<i>Proteus</i>	0	0	1 (0.9)	1 (0.3)
Other		1 (1.0)	0	0	1 (0.3)
Total		97	119	108	324

Table 4 Post-partum distribution of quarter milk samples considering: all quarters, bacteriologically negative quarters, quarters with increased SCC and quarters of cows with increased SCC
Sub-group results according to different inclusion strategies for therapy

Post-partal Diagnosis	No. of quarters (%)																
	All observed quarters No. of quarters (%)						Sub-group results according to different inclusion strategies for therapy										
	HDT	ITS	U	HDT	ITS	U	BAC-	HDT	ITS	U	QSCC100-	HDT	ITS	U	QSCC200-	HDT	ITS
Normal secretion (NORM)	87 (68)	101 (70)	110 (65)	68 (70)	83 (70)	76 (70)	40 (77)	53 (74)	39 (64)	55 (75)	68 (74)	53 (62)	68 (81)	82 (73)	67 (64)		
Increased SCC (SCCI)	17 (13)	17 (12)	21 (15)	12 (12)	15 (13)	16 (15)	5 (10)	7 (10)	9 (15)	7 (10)	10 (11)	15 (17)	8 (10)	11 (10)	18 (17)		
Minor pathogens infection (MIPi)	2 (2)	10 (7)	10 (7)	2 (2)	8 (7)	7 (6)	-	5 (7)	6 (10)	-	5 (5)	7 (8)	-	6 (5)	9 (9)		
Not environment associated major pathogens infection (MAPI)	9 (7)	9 (6)	3 (2)	4 (4)	6 (5)	3 (3)	3 (6)	2 (3)	3 (2)	5 (7)	3 (3)	2 (2)	4 (5)	7 (6)	2 (2)		
Infection with environment associated pathogens (EAPI)	13 (10)	7 (5)	13 (10)	11 (11)	7 (6)	6 (6)	4 (8)	5 (7)	6 (10)	6 (8)	6 (7)	9 (10)	4 (5)	6 (5)	8 (8)		
Total no. of quarters	128	144	136	97	119	108	52	72	63	73	92	86	84	112	104		

indicates proportions of 75% for HDT and 74% for ITS to be detectable with this sample size.

Regarding the main sub-categories there is little change compared to the analysis of pathogens performed in Table 4. There are no significant differences between the groups of pathogen-free sample proportions. Analysis of results on quarters from cows expressing low SCC at the end of lactation provides a different profile. This was performed to meet manufacturer's recommendation to only Teat Seal cows with <200,000 cells/ml. In the ITS group a total of 82 quarters (73% of 112) showed normal secretion after parturition. Adding 11 quarters with elevated cell counts but without bacteriological findings, a total microbial protection rate of 83% was calculated. The Homeopathy group showed a normal secretion rate of 81% (68 of 84) and a microbial protection rate of 91%. For the untreated control the normal secretion proportion was 64% (67 of 104) and infection protection rate 81% (85 of 104). There were no statistical significant effects by the herd concerning the normal secretion rate after calving. The number of cows treated with the eight different remedies was too low to allow any conclusion on the efficacy of particular homeopathic medicines (see Table 5).

The results of the logistic regression models within each treatment group are shown in Table 6. Considering single effects by treatment and udder health status prior to DO and interaction terms of treatment and udder health variables, there was a significant effect in quarters of cows, with less CSCC in animals receiving homeopathy. The OR was 9.69 (95% CI 2.73-34.47) indicating a more than 9-fold chance of protection in these cows. After ITS treatment, there was a non-significant OR of 3.09 (0.86-11.14) in the same sub-group compared to untreated control. A marginally non-significant effect ($p = 0.054$; *post hoc* power 31.3%, OR 18.93 [0.94-380.73]) was detected in quarters infected by minor pathogens after ITS treatment.

SCC of the cows during three milk recording months after calving showed no significant difference between the three treatment groups. A slight decrease in SCC in months 2 and 3 post-partum was observed in all groups (Figure 1a and b).

Discussion

This study was performed to assess efficacy of different antibiotic-free DO treatment methods in comparison to

Table 5 Normal secretion rate after calving of treated udder quarters by homeopathic medicine

Remedy	Treated cows/quarters	Post-partum normal secretion (% quarters)
Calc	10/40	24 (60%)
Calc phos	4/16	13 (81%)
Lach	2/8	8 (100%)
Phos	3/12	9 (75%)
Puls	6/24	18 (75%)
Sep	3/12	9 (75%)
Sil	3/12	4 (33%)
Sulphur	1/4	2 (50%)
Total	32/128	87 (68%)

Table 6 Logistic regression model with dependent outcome variable NORMAL post-partum secretion (<100,000 cells/ml, no pathogens). Final models after non-significant factor reduction ($P > 0.05$) of the full model considering the variables TREATMENT, QSCC, CSCC (cow SCC) and BAC and interaction terms TREATMENT with QSCC, CSCC and BAC (QSCC as not significant omitted)

Variable	Category/interaction term	z	OR	(95% CI)	p
Intercept		0.483	-	-	0.629
TREATMENT	HDT	-1.456	0.19	0.02-1.77	0.145
	ITS	-0.830	0.40	0.04-3.52	0.406
	U (reference)		1		
CSCC	<200,000/ml	-1.30	0.53	0.20-1.38	0.193
	≥200,000/ml (reference)		1		
BAC	Negative	1.15	2.67	0.50-14.36	0.252
	Minor pathogens	-0.346	0.72	0.11-4.56	0.729
	Major pathogens (reference)		1		
TREATMENT × CSCC	HDT × <200,000/ml	3.509	9.69	2.73-34.47	<0.001
	ITS × <200,000/ml	1.728	3.09	0.86-11.14	0.084
TREATMENT × BAC	HDT × negative	0.091	1.11	0.12-9.91	0.928
	HDT × minor pathogens	1.194	4.63	0.37-57.28	0.232
	ITS × negative	-0.020	0.98	0.12-8.01	0.984
	ITS × minor pathogens	1.920	18.93	0.94-380.73	0.055

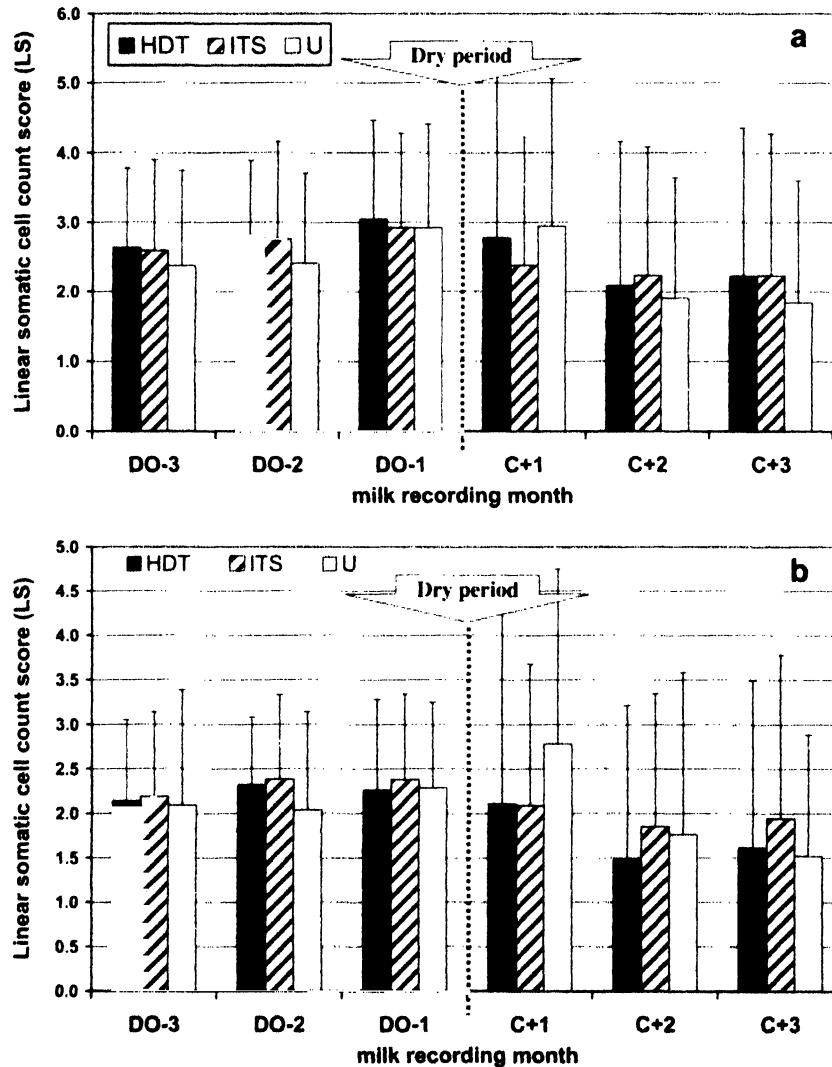


Figure 1 (a and b): Logarithmically transformed SCCs (Linear Score) and standard deviations of cows three months before DO (DO-3 to DO-1) and 3 months after calving (C + 1 to C + 3) in the three treatment groups HDT. (a) All cows, (b) cows with initial cell count <200,000/ml⁻¹. Differences between groups and control dates are not significant ($p > 0.05$).

untreated control. Organic farmers in particular do not require solutions with the gold standard of prophylactic intra-mammary antibiotics because of the regulations to avoid preventive chemical treatment. Antibiotic dry cow treatment is only permitted with veterinary diagnosis and culture of a milk sample. This strategy can be expensive and thus the invention of non-antibiotic devices with no pharmacologic effects such as ETS and ITS was interesting to organic farmers. The problem is the lack of antibiotic protection; this increases the risk of (a) invasion of pathogens with subsequent infection during application and (b) the protection of latent, undiagnosed infections, actually increasing the risk of clinical mastitis. In consequence a SCC threshold 200,000/ml before using these devices at DO is suggested.

The comparison of treatment strategies with antibiotic DCT was not investigated because the blanket use of antibiotics for all cows is not an option on organic farms. Among the different strategies we tested, the Teat Sealing concept seemed to achieve comparable or better results than antibiotics in healthy cows.^{12,13} A recent meta-analysis of antibiotic DCT measures indicates new IMI rates of 6.6–8.0% after antibiotic treatment compared to 12.8% without any therapy.²⁴ In this study the new infection rate in formerly negative quarters, varied from 15 to 18% in the three treatment groups and is higher than in this meta-analysis. Comparison with data of a study which compared antibiotics alone to combined antibiotic and ITS treatment showed new IMI rates of 20% after combination and 25% after only antibiotic therapy until the first three days in milk.¹⁵

Environment associated infections in particular can be prevented by ITS.¹² The present study could not confirm this statistically, but shows tendencies which support these results. Considering all treated quarters, only 5%, compared to 10% in the other treatment groups, were infected by EAPI after the first week of lactation.

None of the strategies investigated here is primarily able to cure sub-clinical mastitis because of the lack of an antibacterial component. The rate of *Staphylococcus* ssp. infections decreased after parturition (11.0% at DO and 5.4% after calving) indicating self cure effects during the dry period, particularly in the homeopathic group where these infections fell from 13 to 1.6%. In contrast, there was only a slight decrease in the untreated control and the Teat Sealer groups. In theory, the therapies ought to be more effective if only culturally negative quarters are considered, but in fact there was little difference in profiles recorded after calving between the categories.

Regarding the recommendation to avoid Teat Sealers as the sole strategy for cows with SCC < 200,000/ml in the last milking record before DO¹²; we took this pre-selection criterion into account and differentiated the protection results of cows above and below this limit at DO. In this trial the proportion of cows exceeding this criterion was 73.5%. Of these 38 quarters (12.7%) were infected by minor and 15 (5%) by major pathogens at DO, with the latter at least having an increased risk of developing post-parturient infection. Of the seven MAPI positive quarters prior to DO treated by Teat Sealers, four were

cured, two showed a major pathogen infection and one a minor pathogen infection. Regarding the spontaneous resolution of most of these cases, a higher risk can not be found for sub-clinical cases treated by Teat Sealers at DO.

Comparing results of the logistic regression model, it can be shown that the homeopathy protocol lead to significant protection effects in cows with SCCs < 200,000/ml in the last milk record prior to DO. This criterion is suggested by the manufacturer of the Teat Sealer as an indicator for treatment. In the ITS group there is a marginally non-significant effect considering this restriction criterion. A cow-level based treatment criterion is transparent for the user and thus practical. Two studies^{12,13} showed positive effects in these sub-groups as well. It can be assumed that missing significance in our study is due to lack of power caused by the small sample size.

In the Teat Sealant group the protection odds are positively influenced by the presence of minor pathogens compared to major pathogens. The presence of minor pathogens at DO time does not increase the risk for mastitis at the start of the subsequent lactation. But it cannot definitely be concluded that minor pathogens offer protection against major pathogens as mentioned by De Vliegher.²⁵

Finally, particularly from the viewpoint of organic farmers, we must discuss which effects can be seen in primarily infected quarters by omitting any therapy. Of the untreated quarters showing sub-clinical infections prior to DO, 36% were still infected after calving. 25% of the quarters were EAPI positive, indicating the tendency for a higher risk with respect to problems by environmental pathogens. But 66% were negative after calving which can be interpreted as self cure/spontaneous resolution during dry period. Regardless of the treatment method bacterial (self) cure rates were: 87, 73, and 33% for the main pathogens *Staphylococcus* ssp., *S. uberis* and *S. aureus*, respectively. Eleven of 12 *Staphylococcus* ssp. positive quarters were bacteriologically negative which raises the question of protection offered by minor pathogen infection of low pathogenicity.^{25,26} Drying MIPI infected quarters hypothetically inhibit the invasion and infection by other microorganisms of higher pathogenicity.

The concept of herd-specific homeopathic treatment does not meet the general principle of individualized homeopathic 'constitutional' therapy, and the standardized application of *Silica 6x* at the end of the therapy cycle is not in accordance to classic homeopathy principles. But this system was developed for inexperienced farmers, using a pragmatic approach which considers the herd as an individual, it is more specific than standardized clinical homeopathy, and avoids the reduction of the remedy determination process in accordance to the disease only. The determination of the herd-specific remedy is part of the farm evaluation process of the extension service in which the farms were participating. The rationale of this procedure can be found in the observation that individuals within a herd show similar behaviour, disease profiles and constitutional traits as well as environmental conditions. The promising effects we found comparing homeopathic remedies to untreated controls permits the hypothesis that considering the individuality of the cows in terms of classical

homeopathic methods might lead to better protective effects against dry period infections.

A problem of this study is the sample size and the lack of statistical power. A similar study design using more cows and farms must be performed to confirm the promising results. Increasing power could show significant effects from Teat Sealants as well. Comparison with long-term antibiotics has not been considered due to prohibition of standard therapy with chemical drugs in organic farming. This study was performed to provide specific solutions for organic farmers.

Conclusion

We conclude that in organic cows with low SCC homeopathy, individualized at herd level and administered at DO, confers highly significantly increases the probability of normal secretion after subsequent parturition, compared to untreated control.

The low incidence of clinical mastitis, which was lowest in the control group, even justifies the abandonment of any therapy in herds in good general condition and reduces environmental impact. The hypothesis that Teat Sealers are an effective prevention measure cannot be excluded on the basis of our data.

Economic analysis should be performed in future, although the results of Berry²⁷ show a cost advantage to treating animals with antibiotics at DO. Considering the fact that these substances in general are undesirable in organic farming, the comparison of the slightly elevated risk of dry period infections in relation to the costs of therapy is required for final strategic decisions.

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