19 January 2017
EMA/CVMP/EWP/444475/2016
Committee for Medicinal Products for Veterinary Use

Overview of comments received on Guideline on the conduct of efficacy studies for intramammary products for use in cattle (EMA/CVMP/344/1999-Rev.2)

Interested parties (organisations or individuals) that commented on the draft document as released for consultation.

Stakeholder no.	Name of organisation or individual
1	Association of Veterinary Consultants (AVC)
2	IFAH-Europe

30 Churchill Place ● Canary Wharf ● London E14 5EU ● United Kingdom
Telephone +44 (0)20 3660 6000 **Facsimile** +44 (0)20 3660 5555
Send a question via our website www.ema.europa.eu/contact

1. General comments – overview

Stakeholder no.	General comment (if any)	Outcome (if applicable)
2	IFAH-Europe welcomes the opportunity to comment on the revised version of the guideline which allows a clearer reading and interpretation and appreciates the consideration given to our comments on the previous version.	

2. Specific comments on text

Line no.	Stakeholder no.	Comment and rationale; proposed changes	Outcome
75-76	2	Please update the reference to the newly adopted revision of the guideline.	Accepted.
119-121	1	**Comments**: We strongly recommend to use GCP as the relevant quality standard for efficacy and clinical studies, while GLP should be acceptable, but not the rule. Therefore propose to modify the wording. **Proposed change**: It is recommended to conduct clinical studies according to Good Clinical Practice (GCP), Good Laboratory Practice (GLP) is also acceptable. In case GCP and/or GLP are not applied, traceability and integrity of data should be adequately guaranteed by other means. For clinical field trials, GCP status is required.	Accepted.
122-142	2	**Comments**: In the CVMP's answer, the position of "well-established use" products according to Art.13a of Directive 2001/82 CE as amended, does not seem to be covered. *Article 13a* states : 1. *By way of derogation from point (j) of the first subparagraph of Article 12(3), and without prejudice to the law on the protection of industrial and commercial property, the applicant shall not be required to provide the results of safety and residue tests or of pre-clinical tests or clinical trials if he can demonstrate that the active substances of the veterinary medicinal product have been in well-established veterinary use within the*	Not accepted. This guideline is intended for cases where new data has to be generated in support of clinical efficacy for products for intramammary use in dairy cattle. Well-established use products according to Art. 13a of Directive 2001/82/EC as amended are not covered. To better address this situation the wording in section 2, Scope, has been modified accordingly. In consequence the change of the wording in section 5.1 (line 131) as proposed, has not been considered.

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		Community for at least ten years, with recognised efficacy and an acceptable level of safety in terms of the conditions set out in Annex I. In that event, the applicant shall provide appropriate scientific literature. » which means that scientific literature could be sufficient to define the dose selection principles. **Proposed change**: Please add at the end of line 131 "Published literature … may be used a supportive information **or pivotal information on a case by case basis**." **Comments**: When administering an intramammary product, the active amounts are directly in contact with the target pathogens and therefore the *in vitro* based- and theoretical PKPD approach is, in the Industry's point of view, all the more relevant when comparing with other systemic administration routes. Consequently the Industry does not understand why a PKPD approach would not be pivotal in setting the appropriate dose but rather would only rely on *in vivo* dose determination studies. This would also be in agreement with the guideline for the demonstration of efficacy for veterinary medicinal products containing antimicrobial substances (EMA/CVMP/627/2001 Rev. 1). Moreover, the requirement for dose determination studies through experimental infections performed with an udder pathogen which is relevant for the claimed indication, is not aligned with the 3Rs	PK/PD approach – not accepted. It is common practice that for intramammary products PK and PD data are used to support the selection of a dose. However, at present there are concerns to base dose finding on a PK/PD approach. Administration of an intramammary product is a local treatment where the dose is not expressed on a body weight basis. Administration and - in the majority of cases - most of the excretion concern the same compartment. There will usually be a large variability between cows (e.g. udder size, level of milk production). *In vivo* the local availability of the active compound is normally assessed by sampling milk and blood. However, it remains unclear to what extent a certain kinetic profile of an active substance in milk correlates with concentrations in the udder tissue. Apart from the difficulties to interpret the PK-profile of a substance there might also be difficulties to interpret the *in vitro*-susceptibility of bovine mastitis pathogens since validated veterinary breakpoints for mastitis pathogens are scarce. Finally it can be noted that currently there is no validated approach for the establishment of a PK/PD relationship which would allow for a waiver of dose determination studies. These are reasons why a reference to a PK/PD approach for

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		principles since often an intramammary product aims to target different pathogens and therefore would involve numerous animals that would be sacrificed. **Proposed change**: Please add at the end of line 134**:** "**A PK/PD approach may waive a dose determination study when justified and confirmed thereafter with a dose confirmation study**."**	dose finding is not given in this guideline. In consequence classical dose determination studies with 3 different dosages and a negative control group are considered indispensable. Taking account of the 3Rs principles this approach is considered justified since mastitis is a very common disease in dairy cattle which requires a sound data base for the dosage regimen.
141-142	1	**Comments**: We propose to offer the use of field studies for any intramammary claims in the absence of experimental models. **Proposed change:** In the absence of experimental models for intramammary prevention and therapy dose determination should be conducted under field conditions.	Accepted. In lactating cows the possibility to study dose determination in naturally infected animals in the absence of experimental models is already included (please, see the last paragraph under the subheading "Experimental studies in lactating cows").
163-164	1	**Comments**: The "internal validity of the study" should be clarified. This sentence is not used in the following section (5.4. Field studies). Is there any specific rule referring to the use of positive control in a dose confirmation study?	There is no specific rule for the use of a positive control in a dose confirmation study. In any trial where a test product is compared to a positive control the internal validity is a particular issue to be considered. It refers to the extent to which one can accurately state that the test product led to the observed effect. That implies that design and implementation of a positive-controlled study needs careful consideration. That could, for example, also include that the results of a positive-controlled study needs to be substantiated by further meaningful data from outside that study.
188-194	1	**Comments**: We agree that any product can be used	Not accepted.

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		as positive control group treatment in a field study that has a marketing authorisation according to Directive 2001/82/EC for the same claim and species as intended to apply for. Regarding the current susceptibility, we would welcome the requirement that MICs of target pathogens are evaluated in any such study, which will allow judgement of susceptibility. As animals are randomised, a reasonable overview for each farm should be available. **Proposed change**: Delete the following sentence: Products for which recent susceptibility data suggest that posology may be inadequate for the infection under study, or products where posology differs between Member States should be avoided. Replace by: for each patient, causative pathogens shall be identified and sensitivity shall be tested for both the active ingredients of the IVP and the control products. An assessment of the potential influence of susceptibility shall be made.	It is agreed that any product that has a marketing authorisation according to Directive 2001/82/EC for the same claim and species can be used as positive control. Nevertheless it is considered advisable to provide a note in the guideline that a reference product should be checked for adequacy which should include the aspects mentioned in the guideline text. The proposal with regard to susceptibility testing is in principle covered by the text in section 5.4.4. The sentence in question which is proposed for deletion has a different tenor which is considered an important advice. Therefore, it will be kept.
197-199	2	**Comments**: Since the identification of the mammary pathogen can only be determined by complementary analysis, most of the time bacteriology on milk sample (results available 24 hours later), to perform a field study with a negative control, in our mind • is not ethical • negatively impacts the inclusion rates into the trial due to owner's reluctance, especially in dairy cattle where genetic value of the animals	It is agreed that the conduct of a negative-controlled clinical trial, which is required for mastitis infections with a high spontaneous cure rate in lactating cows, is usually not acceptable under field conditions. Therefore, for such cases as addressed above it is advised to perform a dose confirmation study under laboratory conditions with a negative control group.

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		is of utmost importance. The industry is aware that infections with high spontaneous cure rate exist but would like to underline that it is an a posteriori consideration. At the moment of the potential inclusion of a lactating cow suffering from clinical mastitis in a trial, the pathogen is not known; thus the potential spontaneous cure cannot be predicted. To postpone the treatment until the identification of the involved pathogen may lead to a significant change in the animal's condition in either a positive way or also in a negative manner which is not ethical. This reduces the chance to cure for the animal which is unacceptable from the farmer's perspective. This is supported by Van den Borne *et al*., in 2010, who reminded "If treatment is delayed, allowing the duration of infection to increase, treatment success seems to deteriorate." Moreover, the CVMP gave *Escherichia coli* infection as an example of high cure rate mastitis and we agree; but mastitis with *E. coli* may also become chronic. Johannes Martinus Swinkel wrote in 2014 in his thesis "Extended antibiotic treatment of persistent bovine mastitis during lactation (Efficacy, economics and social influences)" "*Some bacterial species, such as Escherichia coli, usually show a short transient pattern (De Haas et al., 2004, Schukken et al., 2011). However, dependent on*	

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		the individual host immune response (Burvenich et al., 2003), E. coli may also cause an acute, life threatening, clinical inflammatory response. *[…]* *For example, E. coli usually causes transient infections, but some bacterial strains appear more cow adapted and cause persistent infections (Döpfer et al., 1999, Bradley and Green, 2001, Dogan et al., 2006). An acute clinical inflammatory response to invading mastitis pathogens such as E. coli, may be a concern for dairy farmers as it can be a life threatening condition for the cow."* To conclude, taking into account that: • The pathogen involved in the mastitis is unknown at the moment of inclusion; • Waiting for a bacteriological diagnostic before starting the treatment would lead to an important reduction in the chance of successful treatment (particularly in case of *Klebsiella* infection); • In case of confirmation of mastitis due to *Escherichia coli*, the spontaneous cure character is still unknown. Whilst we acknowledge that negative control data are required when dealing with infections with a high spontaneous cure rate, it should not be made mandatory to generate those data under field conditions. The negative control data originating from	

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		the dose determination study under lab conditions should suffice. For these reasons we are strongly convinced that a negative control in a field trial would be unethical and is not appropriate. References 1/ **Bradley AJ, Green MJ,** 2001. Adaptation of *Escherichia coli* to the bovine mammary gland. Journal of ClinicalMicrobiology 39, 1845-1849. 2/ **Burvenich C, Van Merris V, Mehrzad J, Diez-Fraile A and Duchateau L,** 2003. Severity of *E. coli* mastitis is mainly determined by cow factors. Vet Res. 34:521-64. 3/ **De Haas Y, Veerkamp RF, Barkema HW, Gröhn YT & Schukken YH,** 2004. Associations between pathogen-specific cases of clinical mastitis and somatic cell count patterns. *Journal of Dairy Science* 87 95-105. 4/ **Dogan B, Klaessig S, Rishniw M, Almeida RA, Oliver SP, Simpson K & Schukken YH,** 2006. Adherent and invasive *Escherichia coli* are associated with persistent bovine mastitis. *Veterinary Microbiology* 116 270-82. 5/ **Döpfer D, Barkema HW, Lam TJGM, Schukken YH, and Gaastra W,** 1999. Recurrent clinical mastitis caused by *Escherichia coli* in dairy cows. J Dairy Sci. 82:80-85. 6/ **Schukken YH, Bennett GJ, Zurakowski MJ, Sharkey HL, Rauch BJ, Thomas MJ, Ceglowski B, Saltman RL, Belomestnykh N & Zadoks RN,** 2011.	

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		Randomized clinical trial to evaluate the efficacy of a 5-day ceftiofur hydrochloride intramammary treatment on nonsevere gram-negative clinical mastitis. *Journal of Dairy Science*94 6203-6215. 7/ **Johannes Martinus Swinkel**, 2014. Extended antibiotic treatment of persistent bovine mastitis during lactation (Efficacy, economics and social influences). 8/ **Van den Borne BH, van Schaik G, Lam TJ, and Nielen M,** 2010. Therapeutic effects of antimicrobial treatment during lactation of recently acquired bovine subclinical mastitis: two linked randomized field trials. J Dairy Sci.93: 218-233. **Proposed change**: ~~Comparison with a negative control is also considered necessary for infections with a high spontaneous cure rate (e.g. some subclinical infections, Escherichia coli infections in lactating cows), since a non-inferiority study design is unlikely to yield conclusive information for this situation.~~ **Comparison with a negative control is also considered necessary for subclinical infections**.	
201	1	**Comments**: as explanations on the control are given above, not sure why this sentence is needed. **Proposed change**: delete sentence.	Accepted.
221	1	**Comments**: it should be clearly stated that broth dilution techniques should be used or comparable	Accepted.

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		methods in line with CLSI recommendations. **Proposed change**: according to recognized procedures (e.g. broth dilution methods as recommended by CLSI).	
268	1	**Comments**: The Exclusion criteria subsection refers to the section 5.4. Field studies. Is that applying also for 5.2 and 5.3 (dose determination and dose confirmation)?	For the interpretation of the clinical data package it is reasonable to use the same exclusion criteria.
271-272	1	**Comments**: the 30 day period is to generic and should rather be possible to accommodate according to products used. **Proposed change**: Cows given systemic or intramammary anti-infectious or anti-inflammatory treatments within a period before the trial that may influence the results of treatment of such cow (e.g. a few days in short acting products, longer in case of longer acting products; duration to be justified).	Accepted. The proposed wording is considered sufficient without the examples in brackets.
273	1	**Comments**: this reads like applying for live long period in an animal. However, even if an animal might have been vaccinated 1 year ago, it may develop clinical disease and need treatment. Therefore, this sentence is too broad in its consequence of selection of animals. **Proposed change**: Cows treated with products	Not accepted. It is acknowledged that vaccinated animals may develop a mastitis and need treatment. However, for the purpose of efficacy studies the inclusion of such cows cannot be recommended since it is not known whether there may be an influence on the results of an intramammary treatment of such cow even if the vaccination has been done quite long ago. As far as possible any bias should be avoided.

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		inducing an immune-mediated response against mastitis pathogens may be limited to be enrolled.	
309-10, 311-312	2	**Comments**: A re-infection of an originally infected, treated quarter is possible with a pathogen that is a different bacterial species or that belongs to the same pathogen species, but can be clearly differentiated from the original pathogen. **Proposed change**: Line 309-310: …(absence of the udder pathogen ~~species~~ which was present at the time of inclusion). Line 311-312:…(i.e. detection of an udder pathogen which is **a** different **bacterial species or strain compared to** ~~from~~ that isolated at inclusion …).	Accepted.
328	1	**Comments**: Two positive pre-treatment samples to diagnose a subclinical mastitis is not relevant in case of fluctuating milk excretion of the pathogen (*Staphylococcus aureus*, for example). In such case, only one positive sample should be considered sufficient to include the case in the study.	Not accepted. For the purpose of this guideline there should be strong evidence of infection especially in the field of subclinical mastitis. This is best achieved by the sampling procedure as outlined in the guideline to reach sufficient sensitivity and specificity (i.e. correct detection of infections and non-infections, respectively). Deviations from the sampling procedure for 'cases of fluctuating milk excretion of the pathogen' cannot be supported. It can be noted that this position is supported in a review article concerning *Staph aureus* where it says: "To increase sensitivity of detection of IMI and to account for the fact that shedding of *Staph aureus* may be intermittent, the diagnosis of IMI can be based on culture results of multiple consecutive

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			samples." (Barkema et al. (2006):Invited Review: The Role of Cow, Pathogen, and Treatment Regimen in the Therarpeutic Success of Bovine *Staphylococcus aureus* Mastitis; J. Dairy Sci. 89:1877-1895) Therefore, a modification of the wording is not considered necessary. In addition this requirement is in principle in line with those of the FDA/CVM (i.e. "Prior to treatment, two single microbiology and QSCC samples will be obtained at a 24-hour interval").
88-89 and 278 and 330 - 331	2	**Comments**: Taking into account the above mentioned 3R-principles one should consider to allow inclusion of 2 (and more) udder quarters per cow in case of subclinical mastitis if the detected pathogen is the same in all affected quarters. **Proposed change** (if any): Delete line 278. Line 330-331: More than 1 quarters per cow may only be included if the detected pathogen is the same in all quarters applicable.	Not accepted. It has been decided that only cows with one subclinically infected quarter qualify for inclusion. The reason is that a difference in the response to treatment between cows with one affected quarter and cows with more than one affected quarter have to be taken into consideration for the efficacy evaluation. Moreover, interference between quarters cannot be excluded. Consequently, the option that more than 1 quarter may be included is not supported.
333	1	Same remark: two consecutive samples 1 to 3 days apart might be not relevant for *S. aureus*.	Not accepted. See above.
365	1	Same remark about *S. aureus* infections.	Not accepted. See above.
385	2	**Comments**: Please specify duration of the "colostrum stage" (24/ 48 hrs? up to 5 days?).	Accepted. The colostrum stage has been further specified.

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		Proposed change: The first milk sample should not be taken before 48 hrs (…- tbd) after calving.	
403	2	**Comments**: Please include 'target' for the definition of a prevention success as only infections with target pathogens are of relevance for the definition of a prevention success and not the detection of other udder pathogens for which the product is not indicated. **Proposed change**: ‚…if no **target** udder pathogens can be detected…'	Accepted.
431	1	**Comments**: Using "Combined cure rate" here is confusing since in section 5.4.10 treatment success is based on bacteriological cure. As stated (line 348) "a marked decrease in the SCC is considered as supportive". SCC could remain elevated and take a long time to decrease and is not necessarily linked to the presence of IMI.	Accepted. The respective sentence has been deleted.
447	1	**Comments**: "Clinical trial" to be precised: field trial or is an efficacy laboratory study is possible?	The term 'appropriate clinical trial' is the wording as used in Art. 13 (3) of the Directive 2001/82/EC. To achieve meaningful results with regard to comparable efficacy between a test and a reference intramammary product only a clinical trial under field conditions is considered adequate. The wording in the guideline has been adjusted accordingly to avoid uncertainty.
466	2	**Comments**: In general, excipients used in generic	Accepted.

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		products are very similar to those contained in the pioneer products, are classically used and are well known, thus, local tolerance data should not be needed. **Proposed change**: "Local tolerance data might be requested" could be removed or the cases which require local tolerance data should be specified.	
526-527	2	**Comments**: If there is more than one active substance in the product then the crystalline form of each active substance should be investigated separately. This sentence means that the crystalline form analysis must be performed on each active substance (raw materials) (the active ingredient used in the pioneer product cannot be characterised) and that the results must be compared to the crystalline form obtained in the formulation (after manufacturing process) of the generic product or that the crystalline form of each active ingredient must be analysed in the pioneer and generic product formulation. In this case, it is not easily possible to obtain the crystalline form of each active ingredient in all type of formulation. **Proposed change**: Could you please clarify this sentence?	Comparison of the crystalline form in the finished product between the reference and the generic product is meant here. Chrystalline form here is directly associated with polymorphism. Different polymorphic forms of an active substance usually have different dissolution characteristics. Therefore, a prerequisite for granting a biowaiver is that active substances in generic and reference product have the same crystalline form. This should be investigated (in the finished product) by physical analysis or obtained by other means (e.g. literature).
529-530	2	**Comments**: "Appearance" (coloration for example) is not a criteria that affect the quality and/or the efficacy of the product.	Not accepted. Appearance is considered a useful attribute to demonstrate similarity between the reference and the generic product.

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		Viscosity and density have an influence on the appearance (aspect) of the product. These parameters are discussed in the guideline. Thus, the criteria "appearance" could be removed. **Proposed change**: The pharmaceutical form should be the same~~, and the appearance of the generic and the reference products should be similar.~~	
534-536	2	**Comments**: "If there is more than one active substance in the product then each active substance should be considered separately." This sentence means that the particle size distribution must be performed on each active substance (raw materials) (the active ingredient used in the pioneer product cannot be characterised) and that the results must be compared to the particle size obtained in the formulation (after manufacturing process) of the generic product or that the particle size of each active ingredient in the formulation must be analysed in the pioneer and generic product. In this case, it seems difficult to obtain and distinguish the particle size of each active ingredient in the formulation. **Proposed change**: Could you please clarify this sentence?	Comparison of particle size distribution for the active substance(s) in the finished product between the reference and the generic product is meant here. Particle size distribution of active substance(s) is a very important attribute since it might have influence on the dissolution rate. Therefore, a prerequisite for granting a biowaiver is that active substances in generic and reference product have a similar particle size distribution. This should be investigated by physical analysis (in the finished product) or obtained by other means (e.g. literature).