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PUBLICLY AVAILABLE ASSESSMENT REPORT FOR THE IMMUNOLOGICAL VETERINARY MEDICINAL PRODUCT

Tricholor

PRODUCT SUMMARY

EU procedure number	DE/V/0293/001/MR
Name and pharmaceutical form	**TRICHOLOR** Live vaccine against trichophytosis, Lyophilisate and solvent for solution for injection, for cattle
Applicant	Ceva Tiergesundheit GmbH Kanzlerstraße 4 40472 Düsseldorf Germany
Active substance(s)	*Trichophyton verrucosum*, strain LTF-130, live attenuated (min. 2 x 10^7 - max. 6 x 10^7 CFU/ml)
ATC vetcode	QI 02 A P01
Target species	Cattle
Indication for use	For prophylactic and therapeutic treatment against trichophytosis caused by *Trichophyton verrucosum*, in cattle from one day of age.

PRODUCT INFORMATION

The Summary of Product Characteristics (SPC), the labelling and package leaflet for this immunological veterinary medicinal product (IVMP) are available in the Union Product Database (UPD).

SUMMARY OF ASSESSMENT

Legal basis of original application	Mutual Recognition Procedure application in accordance with Article 32(2) of Directives 2001/82/EC as amended.
Date of completion of the original mutual recognition procedure	27.04.2022
Date immunological veterinary medicinal product first authorised in the Reference Member State (MRP only)	03.12.2021
Concerned Member States (CMS) for original procedure	FR

1. SCIENTIFIC OVERVIEW

The IVMP is manufactured and controlled using validated methods and tests that ensure the consistency of the IVMP released on the market.

The IVMP can be safely used in the target species; the slight reactions observed are indicated in the SPC.

The IVMP is also safe for the user, the consumer of foodstuffs from treated animals and for the environment, when used as recommended. Suitable warnings and precautions are indicated in the SPC.

The efficacy of the IVMP was demonstrated according to the claims made in the SPC.

The overall risk/benefit analysis is in favour of granting a marketing authorisation for this IVMP.

2. QUALITY DOCUMENTATION (physicochemical, biological or microbiological information)

2.A. Product description

The IVMP consists of a lyophilisate and a solvent.

The lyophilisate contains live microconidia of attenuated *Trichophyton verrucosum* cultures (strain LTF 130; min. 2 x 10^7 up to max. 6 x 10^7) and the excipients sucrose and gelatine as stabilizers.

The solvent is a physiological sodium solution composed of sodium chloride and water for injections.

The IVMP is an established pharmaceutical form and its development is adequately described in accordance with the relevant European guidelines.

It is intended for intramuscular injection.

The lyophilisate is filled in either 10 ml (2.5- and 5-ml volumes) or in 20 ml (10 ml volume) type I glass vials. The solvent is filled in glass type I vials (10 ml, 20 ml) and glass type II vials (50 ml). The vials are closed with bromobutyl rubber stoppers and sealed with aluminium caps. The vials and stoppers are sterilized adequately and comply with European Pharmacopoeia.

The choice of the vaccine strain, the formulation and the absence of a preservative is justified.

The selection of the manufacturing process of the active substance and the finished product is explained.

Characterisation of the active substance including the determination of biological properties, purity and identity of the active substance is provided in order to allow suitable specifications to be established.

2.B. Description of the manufacturing method

The IVMP is manufactured fully in accordance with the principles of good manufacturing practice at a licensed manufacturing site.

Process validation data on the IVMP are provided in accordance with the relevant European guidelines.

2.C. Production and control of starting materials

The active substance is live microconidia of attenuated *Trichophyton verrucosum* cultures, an established active substance.

The active substance specification is considered adequate to control the quality of the material. Batch analytical data demonstrating compliance with this specification are provided.

Scientific data are provided and compliance with the Note for Guidance on Minimising the Risk of Transmitting Animal Spongiform Encephalopathy Agents via Human and Veterinary Medicinal Products was satisfactorily demonstrated.

Starting materials of non-biological origin used in production comply with respective pharmacopoeia monographs.

Biological starting materials used are appropriately assessed for the absence of extraneous agents.

The master and working seeds were produced according to the seed lot system as described in the relevant guideline(s).

2.D. Control tests during the manufacturing process

The tests performed during production are described and the results of three consecutive runs, conforming to the specifications, are provided.

In-process control tests are carried out on intermediate stages of manufacture in order to verify the consistency of the manufacturing process and the final IVMP.

A specification was set for each intermediate and the analytical methods are described and validated, if applicable.

2.E. Control tests on the finished product

For all tests, a short description of the techniques for analysing the finished product is provided. The tests and their specifications and limits are justified and are considered appropriate to adequately control the quality of the IVMP.

Satisfactory validation data for each analytical methods are provided, if appropriate.

The tests performed on the final product conform to the relevant requirements and monographs.

Batch analytical data from the proposed production site(s) are provided demonstrating compliance with the determined specification.

The demonstration of the batch-to-batch consistency is based on the results of six batches produced according to the method described in the dossier.

2.F. Batch-to-batch consistency

Full protocols of three consecutive batches of the smallest and three consecutive batches of the largest presentation of the product are provided. They are representative of the routine production, giving the results for all tests performed during production and on the finished product in order to ensure that quality is consistent from batch to batch and to demonstrate conformity with the predefined specifications.

2.G. Stability tests

Stability data on the lyophilisate and the solvent are provided in accordance with applicable European guidelines, demonstrating the stability throughout the shelf life when stored under the approved conditions.

The in-use shelf life of 2 hours of the reconstituted product is supported by the data provided. The recommendations in the product leaflet should be followed.

2.H. Other information

None.

3. SAFETY DOCUMENTATION (safety and residues tests)

3.A. General requirements

The safety data package of this dossier has been collected many years ago for the originator product Trichovac LTF 130.

The safety of the IVMP when administered to the target species, the potential harmful effects (residues in IVMP, substance in foodstuff), the potential serious risk for human beings during product administration and to the environment are adequately described.

All batches used in the clinical trials were manufactured and tested according to the instructions for manufacture and testing described in part 2.

3.B. Pre-clinical studies

The safety of the administration of one maximum single dose, a 10-fold overdose and the repeated administration of one maximum single dose to the target animal was demonstrated in six laboratory studies.

Safety of the target animal below 4 months of age (safety of the repeated administration of one dose and safety of a 10-fold overdose)

Study F-ST/03/96 (1996):

Study aim	To demonstrate the safety of Trichovac LTF 130 in calves following administration of a single therapeutic dose and a single overdose
Animals and application scheme (study groups)	12 calves of two weeks of age • 6 calves were immunised intramuscularly three times with a maximum single dose using the shortest recommended interval of 10 days between the vaccinations • 6 further calves of two weeks of age received a 10-fold overdose intramuscularly
Follow-up	The following safety parameters were evaluated: • Measurement of temperature before each injection, 4 hours after injection and daily for four days after immunisation. • Occurrence of local reactions at the injection site throughout the whole trial period. • Occurrence of systemic reactions throughout the whole trial period.
Results	Single dose: No local or systemic reactions or temperature responses over 1.5 °C above the baseline were observed Overdose: In three animals a bronchial infection was observed for 1 day and in one animal this was accompanied by diarrhoea. The respiratory symptoms were considered due to stress factor as a result of transportation, and indeed bovine trichophytosis infections are not causing respiratory disease. One animal (02961) showed an increase in body temperature of 1.6 °C on the first day after vaccination. No local reactions were observed.
Conclusion	The vaccine is safe for calves when the basic maximum therapeutic dose is administered. The vaccine is safe for calves when ten times the maximum usual dose is administered.

Russian laboratory study (1983):

Study aim	To verify the tolerability of vaccination with Trichovac LTF 130 in calves of different ages
Animals and application scheme (study groups)	46 calves aged from 4 to 45 days were vaccinated twice intramuscularly with 1 ml at an interval of 14 days.
Follow-up	All animals were clinically controlled 3 and 24 hours post vaccination, 14 and 56 days post vaccination
Results	No local or systemic reactions were observed 3 and 24 hours post vaccination. No abnormal reactions could be revealed (only typical crusty patches were detected) 14 and 56 days post vaccination.
Conclusion	The vaccine is tolerated by calves in the event of repeat administration of a vaccine dose 14 days after the first. Intolerance reactions have not been reported in any of these cases. Study can be considered as supportive.

Safety for the target animal above 4 months of age (safety of the administration of one dose)

Study F-ST/05/96 (1996):

Study aim	To demonstrate the safety of Trichovac LTF 130 in calves following administration of the basic therapeutic dose
Animals and application scheme (study groups)	6 calves of four months of age were vaccinated intramuscularly with the maximum single dose.
Follow-up	The following safety parameters were evaluated: • Measurement of temperature before each injection, 4 hours after injection and daily for four days after immunisation. • Occurrence of local reactions at the injection site throughout the whole trial period. • Occurrence of systemic reactions throughout the whole trial period.
Results	No local or systemic reactions or temperature responses over 1.5 °C above the baseline were observed.
Conclusion	The vaccine is tolerated by calves when the basic maximum usual dose is administered.

Safety in pregnant animals (safety of the repeated administration of one dose and safety of a 10-fold overdose)

Study F-ST/04/96 (1996):

Study aim	Examine the safety for pregnant cows
Animals and application scheme (study groups)	6 pregnant cows were vaccinated intramuscularly three times with 4 ml (therapeutic dose) at an interval of 10 days. 6 further pregnant cows received a 10-fold overdose. 12 pregnant cows served as control animals to compare the fertility data.
Follow-up	The following safety parameters were evaluated: • Measurement of temperature before each injection, 4 hours after injection and daily for four days after immunisation. • Occurrence of local reactions at the injection site throughout the whole trial period. • Occurrence of systemic reactions throughout the whole trial period.
Results	No systemic reactions or temperature responses over 1.5 °C above the baseline were observed.

	After the 1st administration no local reactions were observed but after the 2nd and 3rd vaccination as well as after the overdose swellings were observed that disappeared within 31 days without treatment. These swellings were seen in adequately sensitised animals and are considered to be a delayed allergic reaction. All animals delivered a healthy calf at term.
Conclusion	The vaccine is safe for use in pregnant cows when • the basic maximum usual dose is administered, • ten times the maximum usual dose is administered, • the basic maximum usual dose is repeatedly administered.

Dissemination (faeces, urine, muscle and skin scrapings)

Study F-ST/12/96 (1996):

Study aim	Examine the persistence and excretion of the vaccine strain and spread of the vaccine strain in four-week-old calves
Animals and application scheme (study groups)	6 calves of 4 weeks of age were vaccinated twice with an interval of 14 days. 2 further calves were used as in-contact control animals, which were not vaccinated and housed together with the vaccinated calves.
Follow-up	The following safety parameters were evaluated: • Presence of the vaccine organism in urine and faeces samples collected at 6 occasions during 14 days after the 2nd vaccination. • Presence of the vaccine organism in muscle biopsies of the vaccinated animals collected at 3 occasions during 14 days after the 2nd vaccination. • Presence of the vaccine organism in skin scrapings of the vaccinated animals, collected at 18 days post the 2nd vaccination.
Results	The vaccine organism could not be found in urine, faces and muscle samples. There was one positive urine sample of a control animal on the day of 2nd vaccination, which is considered an artefact due to cross contamination. The vaccine strain could be reisolated in 3 out of 6 skin scrapings taken from the crusty patches at the injection site.
Conclusion	The vaccine strain does not disseminate in the vaccinated animals, is not excreted in urine and faeces and does not spread to calves that are in direct contact, even not if skin scapings remain positive for some time.

Dissemination (milk)

Study F-ST/04/97 (1997):

Study aim	Examine the persistence and excretion of the vaccine strain and spread of the vaccine strain in lactating dairy cows
Animals and application scheme (study groups)	6 lactating cows were vaccinated twice with an interval of 14 days. 2 further lactating cows were used as in-contact control animals, which were not vaccinated and housed together with the vaccinated cows.
Follow-up	A total of 48 milk samples were collected at 6 occasions during 14 days after the 2nd vaccination (6 samples per cow).
Results	The vaccine strain could not be found in any milk sample.
Conclusion	The vaccine strain does not pass into the milk when a repeat maximum therapeutic dose of the vaccine is administered.

Specific studies were carried out to describe the spread and dissemination of the vaccine strain. The vaccine strain is not excreted from vaccinated animals and thus, it does not spread. This is confirmed by the fact that the naive in-contact control animals in both studies did not get infected, even not if they were housed together.

There are no data suggesting that this product might adversely affect the immune system of the vaccinated animal or its progeny. Therefore, no specific study was carried out.

Reversion to virulence, biological properties and recombination or genetic reassortment of the vaccine strain was evaluated. It was concluded, that increase in virulence due to animal to animal passage can be excluded and genetic recombination as the basis for virulence reversion should not be regarded as a realistic risk. No warnings were needed in the SPC.

As the vaccine contains a zoonotic agent it is obvious that there is some occupational risk related to the use of this vaccine. Therefore, a warning is included in the SPC section 3.5 that the use of gloves is recommended and that a physician should be consulted upon accidental self-injection and that the leaflet/label should be shown.

For none of the excipients used an MRL applies. Based on this information and on the information about dissemination, no withdrawal period is necessary for eggs. For meat and offal the withdrawal period was set at 7 days, as the vaccine contains live microconidia, which must be present for a while at the injection site. The biopsies of muscles started 7 days after injection, being negative. So, it was demonstrated that no live microconidia were reisolated 7 days after immunisation. Having in mind the zoonotic potential of the vaccine strain the withdrawal period for meat and offal was set at 7 days, accordingly.

No specific assessment of the interaction of this product with other medicinal product was made. Therefore, an appropriate warning in the SPC is included.

3.C. Clinical trials

The reports of four field trials conducted in Germany are provided.

In addition supporting literature data are provided (Holubek et al., 2000).

Trial in Griepentrog dairy herd (1994-1995):

Study aim	The aim was to investigate the safety of Trichovac LTF 130 in a field scale trial.
Animals and application scheme (study groups)	The entire herd was vaccinated twice with a therapeutic dose (2 x 2 ml or 2 x 4 ml intramuscularly) including: • 229 cows (not pregnant) • 669 inseminated / pregnant cows in all stages of pregnancy • 349 inseminated / pregnant heifers in all stages of pregnancy • 181 calves aged from 3 days to 4 months • 819 young female cattle from 4 months of age In the further course ongoing vaccinations were performed on all calves from three days of age; at the beginning with the therapeutic dose (2 x 2 ml) due to the high infection pressure, later on with the prophylactic dose (2 x 1 ml) using an interval of 14 days.
Follow-up	• Recording of local and general vaccine reactions • Observation of the influence of vaccination on reproductive performance and milk yield; particularly close observation of animals in the last gestation trimester during the first 12 hours post vaccination
Results	No local or systemic reactions were observed. There was no direct impact of herd vaccination on herd milk yield. The rates of conception and pregnancy as well as the proceeding of pregnancies were not negatively impacted. Abortions and preterm deliveries were not observed in context with the vaccination. The deliveries, post-partum phases and lactation periods were not negatively affected, as well as the development and vitality of the calves.

	The ongoing vaccinations with the prophylactic dose in calves from three days of age did not cause any local or systemic reactions.
Conclusion	The therapeutic dose of the vaccine is tolerated locally and generally throughout pregnancy by all ages, including calves from three days of age as well as cows and heifers. A negative effect on reproduction and lactation was not detected.

Trial in Thumsenreuth dairy herd (1995)

Study aim	To examine the safety of Trichovac LTF 130 vaccine under practical conditions
Animals and application scheme (study groups)	In this herd (200 animals) 12 infected young cattle were vaccinated twice with a therapeutic dose (2 x 4 ml, intramuscularly) 14 days apart. 8 control animals, which were initially unaffected, received the prophylactic dose (2 ml) at the first vaccination. Later on 4 of these control animals showed clinical signs as they already were in the incubation phase at first vaccination. These 4 diseased animals received a therapeutic dose at second vaccination, the 4 unaffected animals were vaccinated with a prophylactic dose.
Follow-up	Recording of local and general vaccine reactions
Results	No local or systemic reactions were observed in the vaccinated animals regardless of the dose.
Conclusion	The vaccine is well tolerated.

Trial in Griebo young cattle unit (1994)

Study aim	To examine the safety of Trichovac LTF 130 vaccine under practical conditions
Animals and application scheme (study groups)	The whole herd (290 heifers) was vaccinated twice with a therapeutic dose (2 x 4 ml, intramuscularly) 14 days apart.
Follow-up	Assessment of the safety of the vaccine Assessment of the impact of vaccination on the performance
Results	No local or systemic reactions were observed. No impact on the performance of the vaccinated animals could be detected.
Conclusion	The vaccine was safe in heifers at the double therapeutic dose of 4 ml.

Trial in Radis cattle herd (1993-1994)

Study aim	To examine the safety of Trichovac LTF 130 vaccine under practical conditions
Animals and application scheme (study groups)	Around 150 young cattle, 270 cows and heifers and 35 calves were vaccinated. Generally, healthy, at risk and diseased young cattle, heifers and cows received 2 x 4 ml and calves 2 x 1 ml intramuscularly. Some heifers and cows were vaccinated 3 times and 2 cows even 4 times due to severe trichophytosis infection.
Follow-up	Assessment of local and general safety. Assessment of influence on pregnancy, birth and post-partum phase.
Results	No systemic reactions were observed. Local reactions could be revealed in 20 % of the cows consisting of small swellings at the injection site, which reached the size of the palm of a hand in individual animals. These swellings subsided after 14 days, in some cases after four weeks at the latest, without treatment. Abortions were not recorded following vaccination (this also applies to the dry cows and heifers that were vaccinated in the last gestation trimester). Pregnancies, deliveries and post-partum phases proceeded entirely normal.
Conclusion	The vaccine is well tolerated.

Conclusion:

The data from the field trials support the results of the laboratory studies and clearly show that the vaccine is safe for the target species if used as recommended in the product literature.

Pharmacovigilance data:

Pharmacovigilance data collected as part of the PSUR cycle as valid for the registration in Germany since March 2009 to February 2021 are provided, showing that the vaccine is highly effective.

Based on the PSUR data it can be concluded that the vaccine is safe and effective when used as appropriate in the target species (e.g. for the last period the incidence of adverse events was 0.00148 %).

3.D. Environmental Risk Assessment

The applicant provided a first phase environmental risk assessment in compliance with the relevant guideline, which showed that no further assessment is required. The assessment concluded that the environmental risk is considered effectively zero are therefore no control measures are required.

Warnings and precautions as listed in the product literature are adequate to ensure safety to the environment when the product is used as directed.

3.E. Assessment required for veterinary medicinal products containing or consisting of genetically modified organisms

Not applicable.

4. EFFICACY DOCUMENTATION

4.A. General requirements

The efficacy data package of this dossier has been collected many years ago for the originator product Trichovac LTF 130.

All batches used in the clinical trials were manufactured and tested according to the instructions for manufacture and testing described in part 2.

4.B. Pre-Clinical Studies

The efficacy of the product was demonstrated in laboratory studies under well-controlled conditions in accordance with the relevant requirements, which show that also very young calves from 3 days of age can be vaccinated and are able to build a protective immunity. Basic requirement for building a protective immunity is the post-vaccinal reaction (crusty patch) at the injection site.

Laboratory trials in calves (< 4 months)

Three dose finding studies were conducted:

Experimental exposure of calves in different age groups following immunisation with graded doses of Trichovac LTF 130 (1984):

Study aim	To find the dose that produced immunity. Furthermore, the aim was to clarify the age from which calves react to vaccination by developing robust immunity
Animals and application scheme (study groups)	167 calves aged from 3 to 45 days were vaccinated twice 10 to 14 days apart with a prophylactic dose of different concentrations (5 groups). 9 animals served as unvaccinated controls.
Challenge	The challenge was done 45-69 days after the second vaccination using a virulent culture of strain *T. verrucosum* 153.
Follow up / evaluation criteria*	The calves were clinically controlled from 7 to 42 days post challenge infection • Constant observation of the animals by a veterinarian • Recording of the response to immunisation as well as to the infection
Results	After 14-16 days, all 9 control animals developed trichophytic lesions with a diameter between 5 to 10 cm and secondary lesions in the areas of head and neck. In the vaccinated animals only a specific allergic reaction developed at the infection site. This reaction started around 7-12 days post infection and had disappeared by day 32 in all vaccinated animals.
Conclusion	The results show that even very young calves from 3 days of age are able to have a protective immune response. Based on the overall picture the dose of 2×10^7 was considered as the minimum dose as in this dose post vaccinal safety and efficacy were found best united.

Experimental exposure of calves following immunisation with graded doses of Trichovac LTF 130 vaccine and study of the effect of a fungicidal ointment applied to the vaccination site (1985):

Study aim	To find the dose leading to immunity and to clarify the influence of local fungicidal treatment at the vaccination site on vaccination success

Animals and application scheme (study groups)	87 calves aged 1-2 months were vaccinated twice 14 days apart with different concentrations of microconidia (9 groups). 6 animals served as unvaccinated controls. To determine the relevance of a vaccinal lesion (crusty patch) for the development of the immunity and thus efficacy of the vaccine, in each group several calves were treated post vaccination with an anti-trichophytosis ointment immediately after vaccination and every seven days thereafter.
Challenge	Two months after the vaccination all calves were challenged with a virulent culture of *T. verrucosum* 153.
Follow up / evaluation criteria*	• Recording of the formation of post-vaccine lesion • Regular examination of animals
Results	The results show that the most animals treated with fungicidal ointment developed no vaccinal lesions, like the control animals, and were not protected against challenge. They became sick following infection. All control animals that were not immunised diseased without exception following experimental infection with trichophytosis. In case of the animals that were not treated with fungicidal ointment, the development of lesions depended on the vaccine dose. Lesions were routinely observed upwards of a dose of 5×10^5.
Conclusion	The local post- vaccine reactions are a measure of the immunogenicity of the vaccine and the development of robust immunity. The treatment of vaccinated animals with fungicidal or fungistatic agents prevents the development of immunity and is therefore contraindicated. Prophylactic efficacy of Trichovac LTF 130 is achieved from a vaccination dose of $2 \times 5 \times 10^5$ microconidia/ml. The formation of a vaccination scab and the number of live microconidia/ml of vaccine are criteria for assessing the efficacy of Trichovac LTF 130.

Experimental exposure of prophylactically vaccinated calves in various age groups (1983):

Study aim	To examine the extent to which calves of different ages develop robust immunity following immunisation with Trichovac LTF 130
Animals and application scheme (study groups)	46 calves aged from 4 to 45 days (10 calves were between 4 and 9 days old) were vaccinated twice intramuscularly with 1 ml (2.5×10^7 CFU/ml) at an interval of 14 days. 6 animals served as unvaccinated controls.
Challenge	61 days after the second immunisation 21 vaccinated animals and 3 controls were challenged with virulent *T. verrucosum* 153 (the other animals were kept for demonstration the Duration of Immunity and were challenged at a later time point)
Follow up / evaluation criteria*	• Recording of the formation of local post vaccine lesions • Regular clinical examination of the animals
Results	21 days after infection. After 28-32 days the skin in the area of the infection site was completely free of crusty patches. The 3 challenge control calves all developed disease with secondary lesions on the head and neck between day 30 and 45. The diagnosis was confirmed by mycological examination. The results of this study support the efficacy of a prophylactic dose (given twice, i.m., 14 days apart) in calves.
Conclusion	Even very young calves vaccinated from 4 days old develop robust immunity after administration of 2×1 ml (2.5×10^7 microconidia/ml) of Trichovac LTF 130. The development of immunity is accompanied by a typical local vaccination reaction.

Conclusion:
It is shown that the chosen minimum prophylactic dose of 2.0 x 10^7 live microconidia/ml induces a robust immunity even in very young calves from 3 days of age showing a good safety profile.
The infection model proved to be effective.
The local treatment of vaccinated animals with fungicidal of fungistatic agents at the injection site prevents the development of immunity and is therefore contraindicated. A respective warning is included in the product literature.

Laboratory trials in young cattle

Study F-ST/16/96 (1996-1997):

Study aim	To demonstrate efficacy in young cattle in accordance with the requirement of Directive 92/18 to demonstrate efficacy in each category of the target species.
Animals and application scheme (study groups)	6 young cattle at 4 months of age were immunised twice intramuscularly 14 days apart with a prophylactic dose (2 ml, 4 x 10^7 CFU per dose). Further 4 animals served as unvaccinated controls.
Challenge	6 weeks after the second vaccination all animals were challenged with a *T. verrucosum* strain isolated from a cow that developed the disease spontaneously (strain E 1487/96), besides two unvaccinated controls, which only were scarified.
Follow up / evaluation criteria*	Clinical inspection of the trial and control animals in terms of ▪ Occurrence of local reactions ▪ Occurrence of systemic reactions
Results	First dermatological reactions at the injection site were seen in individual immunised animals from 14 days post infection, in all vaccinated animals before day 18 post infection. The lesions had completely healed between day 42 and 56 after infection with full hair regrowth. In the control animals, dermatological reactions were found from day 18 post infection and increased until approx. 30 days post infection. Bald patches formed, which were rough to the touch and crusty, and showed the typical clinical picture of ringworm. Generalisation was not observed. Complete recovery and hair regrowth did not take place until day 100 post infection. The control animals that were only scarified showed no signs of disease. Wounds had healed completely after approximately 21 days.
Conclusion	The prophylactic efficacy for young cattle has been demonstrated.

Onset of immunity

Phase challenge infection in calves in different age groups following prophylactic immunisation (1984-1985)

Study aim	To determine the point at which robust immunity develops following prophylactic immunisation
Animals and application scheme (study groups)	90 calves aged between 3 and 35 days were vaccinated twice intramuscularly 14 days apart with a prophylactic dose (1 ml, 3.4 x 10^7 CFU/ml). Further 9 animals served as unvaccinated controls. The calves were divided into three groups according their age (30 animals per group).
Challenge	The challenge was done with a virulent *T. verrucosum* 153 culture in always 5 animals of each group on different points in time after the first vaccination (1, 7, 14, 21, 28, 35 days post vac.).
Follow up / evaluation criteria*	The animals infected were examined regularly for a period of 45 days. Mycological examination to ascertain healing of clinical findings.
Results	Number of diseased calves after experimental challenge infection:

	<table><tr><th rowspan="2">Group</th><th colspan="6">Point of infection after first vaccination in days</th></tr><tr><th>1</th><th>7</th><th>14</th><th>21</th><th>28</th><th>35</th></tr><tr><td>1</td><td>5/5</td><td>5/5</td><td>5/5</td><td>4/5</td><td>0/5</td><td>0/5</td></tr><tr><td>2</td><td>5/5</td><td>5/5</td><td>5/5</td><td>3/5</td><td>0/5</td><td>0/5</td></tr><tr><td>3</td><td>5/5</td><td>5/5</td><td>5/5</td><td>4/5</td><td>0/5</td><td>0/5</td></tr></table> All animals infected on day 1, 7 and 14 after the first immunisation, as well as all control animals diseased with typical signs. The diagnosis was confirmed by mycological examination. 21 days after the first immunisation individual animals were already protected. A reliable protection was shown in all calves infected 28 days after the first vaccination (corresponds to 14 days after the second vaccination). These results occurred in all 3 groups regardless of the age of the animals.
Conclusion	Following prophylactic immunisation of calves with 2 x 1 ml of the vaccine 14 days apart, there is reliable protection against a challenge infection from day 28 after the first immunisation.

In another study (Medexport, 1981) among 30 one-months old calves, these animals developed stable immunity, following double intramuscular vaccination with Trichovac LTF 130, between 21 and 30 days after the second immunisation, while all the control calves became sick.
The vaccinated animals were challenged 7, 10, 14, 21 and 30 days after the 2nd vaccination. 14 days after the 2nd vaccination only one of three challenged animals was protected. 21 and 30 days after the 2nd vaccination all animals had protective immunity whereas all control calves diseased.

Onset of immunity – evaluation of field trials:

The onset of immunity after therapeutic use is shown in the four field trials.
Griepentrog: an improvement was recognized at the second vaccination. 3 months after the first vaccination no clinical symptoms were shown.
Thumsenreuth: approx. 4 weeks after the first vaccination diseased calves showed signs of recovery.
Griebo: an improvement was recognized at the second vaccination. From 2 until 4 weeks after the second vaccination the clinical symptoms recovered until complete healing.
Radis: 3 to 4 weeks after the second vaccination the diseased animals show a clear tendency for healing.

Conclusion:

Overall, the results support an onset of immunity within 4 weeks after the 2nd vaccination, as stated in the product literature.

Duration of immunity

Experimental challenge infection two years after prophylactic immunisation of calves in different age groups (1983-1985)

Study aim	The challenge infections should provide information on duration of immunity
Animals and application scheme (study groups)	46 calves aged from 4 to 45 days (10 calves were between 4 and 9 days old) were vaccinated twice intramuscularly with 1 ml (2.5×10^7 CFU/ml) at an interval of 14 days. 6 animals served as unvaccinated controls.
Challenge	20 vaccinated animals and 3 controls were challenged two years after the second immunisation with virulent *T. verrucosum* 153.

	(21 vaccinated animals and 3 controls were challenged with virulent *T. verrucosum* 153 61 days after the second immunisation during dose finding studies.)
Follow up / evaluation criteria*	The animals infected were examined regularly for a period of 45 days.
Results	The immunised animals were found protected, whereas all three control calves developed a severe form of trichophytosis.
Conclusion	In conclusion double vaccination with a prophylactic dose 14 days apart provides at least two years of protection against trichophytosis.

Duration of immunity – evaluation of field trials and references

Field trial Radis (1993-1994):
In the Radis cattle herd trichophytosis was diagnosed. The disease spread quickly through the herd and clinically affected young cattle, cows and heifers. It was observed that, without exception, cows that had been bought in as pregnant heifers from the Vockerode cattle rearing facility did not become infected. These cows had been prophylactically vaccinated with Trichovac LTF 130 at the age of 8 to 12 weeks, and still showed stable protection against severe infection risk at the age of 6 years and older.
It can be concluded that prophylactic immunisation of cattle with Trichovac LTF 130 confers long-lasting, possibly lifelong immunity.

Literature (Sarkisov, 1987; Medexport, 1981):
Phased controlled challenge were carried out in groups of cattle that had been vaccinated with the liquid vaccine at the age of one to four months (after 1, 3, 5, 8, 12 and 14 months, and after 2, 3, 4, 5, 6 and 8 years).
Whilst manifestations of disease occurred in the non-vaccinated animals after 20 to 30 days, stable protection could still be observed in vaccinated animals eight years after vaccination.
These study results were confirmed after development of the lyophilised vaccine.
The robustness of the immunity of calves to infection was also tested in this context in the former USSR using cultures from different geographical regions as well as from other countries.
The animals immunised with Trichovac LTF 130 showed stable immunity when exposed to virulent strains of different geographical origin and did not develop trichophytosis.

Conclusion:

The data provided from the laboratory study, one field study and the literature in principle support that an immunity over several years, in many cases life-long, is developed.

However, the applicant has only own data from a laboratory study, which support a duration of immunity of two years. Therefore, a duration of immunity of 2 years is stated.

Effect of MDA

No specific study on the effect of MDA was carried out.

However, in the publication presented in annex 13 (Rybnikar et al., 2001), it is shown that cattle that is vaccinated during pregnancy are well protected against challenge after parturition. However, this immunity in the dam is not transferred to the calves as shown by the fact that all calves were equally susceptible to a challenge, irrespective of whether they were obtained from vaccinated or control cows. Although this study was performed with a different but similar vaccine, it can be concluded that:

- In the respective study, it was shown that calves born from vaccinated heifers were not protected and thus there is no transfer of maternal immunity (it is generally assumed that protection is cell mediated rather than antibody mediated).
- The vaccine has therapeutic effects, hence even active immunity from a challenge infection does not block the activity of the vaccine.
- The vaccine is only used on contaminated premises but still the vaccine induces good immunity when given from 3 day of age onwards.

No interference on the efficacy of the vaccine by MDA is to be expected.

4.C. Clinical trials

Efficacy of vaccination was also demonstrated under field conditions in four controlled field trials conducted in Germany.

Trial in Griepentrog dairy herd (1994-1995):

Study aim	The aim was to investigate the efficacy of Trichovac LTF 130 in a field scale trial.
Animals and application scheme (study groups)	The entire herd was vaccinated twice with a therapeutic dose (2 x 2 ml or 2 x 4 ml intramuscularly) including: • 229 cows (not pregnant) • 669 inseminated / pregnant cows in all stages of pregnancy • 349 inseminated / pregnant heifers in all stages of pregnancy • 181 calves aged from 3 days to 4 months • 819 young female cattle from 4 months of age In the further course ongoing vaccinations were performed on all calves from three days of age; at the beginning with the therapeutic dose (2 x 2 ml) due to the high infection pressure, later on with the prophylactic dose (2 x 1 ml) using an interval of 14 days.
Follow-up	Recording of clinical manifestations of trichophytosis in the herd
Results	Already at the 2nd vaccination there was an improvement of the clinical situation of cows and heifers. Four weeks after the 2nd vaccination crusty scabs could be observed only in single animals. Three months after the first vaccination there were no clinical symptoms. In calves, the occurrence of clinical symptoms of trichophytosis was prevented due to the vaccination. The ongoing vaccinations of all (also new bought-in) animals prevented further outbreaks of the disease.
Conclusion	It was shown that the infection was contained and gradually the animals recovered from the severe infection and trichophytosis was eliminated from the herd within 3 months of the first vaccination. This clearly supports the therapeutic claim for the vaccine. Vaccinating calves as early as possible from 3 days old is recommended in order to keep the herd free of trichophytosis

Trial in Thumsenreuth dairy herd (1995)

Study aim	To examine the efficacy of Trichovac LTF 130 vaccine under practical conditions
Animals and application scheme (study groups)	In this herd (200 animals) 12 infected young cattle were vaccinated twice with a therapeutic dose (2 x 4 ml, intramuscularly) 14 days apart. 8 control animals, which were initially unaffected, received the prophylactic dose (2 ml) at the first vaccination. Later on 4 of these control animals showed clinical signs as they already were in the incubation phase at first vaccination. These 4 diseased animals received a therapeutic dose at second vaccination, the 4 unaffected animals were vaccinated with a prophylactic dose.
Follow-up	Recording of clinical manifestations of trichophytosis in the herd

Results	Approx. 2 weeks after the 2nd vaccination diseased calves showed first signs of recovery. The ongoing vaccinations prevented further outbreaks of the disease.
Conclusion	The vaccine is also therapeutically effective in the presence of high infection risk, as demonstrated by new clinical cases in the existing herd. The vaccination of only individual groups of animals did not prove advantageous. Only the consistent vaccination of all offspring achieved the intended outcome and prevented new outbreaks in the herd.

Trial in Griebo young cattle unit (1994)

Study aim	To examine the efficacy of Trichovac LTF 130 vaccine under practical conditions
Animals and application scheme (study groups)	The whole herd (290 heifers) was vaccinated twice with a therapeutic dose (2 x 4 ml, intramuscularly) 14 days apart.
Follow-up	Recording of clinical manifestations of trichophytosis in the herd
Results	Already at the 2nd vaccination some animals showed a decrease of hyperkeratosis. From 2 until 4 weeks after the second vaccination the clinical symptoms recovered until complete healing.
Conclusion	Double vaccination of a herd clinically infected with trichophytosis with a therapeutic dose is effective in promoting the clinical recovery of the animals.

Trial in Radis cattle herd (1993-1994)

Study aim	To examine the efficacy of Trichovac LTF 130 vaccine under practical conditions
Animals and application scheme (study groups)	Around 150 young cattle, 270 cows and heifers and 35 calves were vaccinated. Generally, healthy, at risk and diseased young cattle, heifers and cows received 2 x 4 ml and calves 2 x 1 ml intramuscularly. Some heifers and cows were vaccinated 3 times and 2 cows even 4 times due to severe trichophytosis infection.
Follow-up	Recording of clinical manifestations of trichophytosis in the herd
Results	3 to 4 weeks after the second vaccination the diseased animals show a clear tendency for healing, whilst the pathogen was still detectable. The results show that a vaccination by group is not suitable to prevent a dissemination of trichophytosis in a herd. A herd vaccination with ongoing vaccinations of all new (also bought-in) animals is recommended. This can prevent new outbreaks.
Conclusion	The effectiveness of the therapeutic use was demonstrated but also that prophylactic vaccination is necessary to prevent infection of animals that are still free from the infection. The absence of the disease in animals that were previously vaccinated (on a different farm) is indicative for lifelong protection.

Conclusion:

The data from the field trials support the results of the laboratory studies and clearly show that the vaccine is effective in the target species if used as recommended in the product literature.

Pharmacovigilance data:

Pharmacovigilance data collected as part of the PSUR cycle as valid for the registration in Germany since March 2009 to February 2021 are provided, showing that the vaccine is highly effective.

Based on the PSUR data it can be concluded that the vaccine is safe and effective when used as appropriate in the target species (e.g. for the last period a total of 7 lack of efficacy events in 269 animals were reported – estimated number of treated animals 1888905).

5. OVERALL CONCLUSION AND BENEFIT-RISK ASSESSMENT

The data submitted in the dossier demonstrate that when the product is used in accordance with the Summary of Product Characteristics, the risk benefit profile for the target species is favourable and the quality and safety of the product for humans and the environment are acceptable.

POST-AUTHORISATION PROCEDURES

The SPC and package leaflet may be updated to include new information on the quality, safety and efficacy of the veterinary medicinal product. The current SPC/labelling/package leaflet is/are available in the Union Product Database (UPD).