EXPERIMENTAL EVALUATION OF THE RapidVet™-D TEST

Test for the Rapid Identification of Dermatophyte Infection in Dogs, Cats and Horses

Extract of a summary of a more extensive text submitted for publication.

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INTRODUCTION

Dermatophytes are very important agents of zoonosis because man can contract the infection from all the species of dermatophytes found on animals. The diseases caused by dermatophytes are known as "ringworm" because of the ring-like appearance of the lesion. Dermatophytes have the capacity of metabolizing the keratin on which they feed, so they grow on the keratinized follicle. The most common causes of dermatomycosis are fungi belonging to the genera Microsporum, Trichophyton and Epidermophyton. These fungi are classified, on the basis of their natural habitat, as Geophylous, Zoophylous, or Anthropophylous.

Geophylous dermatophytes (*M. gypseum, M. nanum*) live in the soil. Animals are infected upon contact. Zoophylous dermatophytes (*M. canis, M. distortu, T. gallinae, T. verrucosum* and *T. equinox*) are found on the skin of animals, especially young animals. The expansion of the infection is enhanced by the naturally occurring moisture and temperature of the animal's skin, and by the existence of a wound, abrasion or skin problems caused by poor nourishment. Anthropophylous dermatophytes (*T. mentagrophytes, T. tonsurans, M. audoninii, T. rubrum*) are the primary parasites in man and can live in soil for only very short periods.

As cases of dermatophytosis are clinically similar to many other skin diseases, the diagnosis should be confirmed before beginning antifungal treatment. Several methods have traditionally been employed to identify dermatophytes, including direct microscopic examination of dermal scabs and hair, examination using Wood's lamp under ultraviolet light in a dark room, and examination of an actual culture grown in a controlled environment for up to 14 days. For the veterinarian needing swift confirmation of a diagnosis so that appropriate treatment can be initiated, these methods of examination can be less than ideal.

A decade ago, Taplin and colleagues developed a culture medium for the isolation and identification of dermatophytes. This method is based on the use of a highly sensitive culture medium which allows the growth of dermatophytes (pathogenic fungi) but not saprophytes (non-pathogenic fungi). In order to obtain a yes/no result, a system for color change (from yellow to red) was incorporated into the medium which reacts in the presence of metabolites produced by dermatophytes. Because certain bacteria and saprophytic fungi can grow even on this selective medium, three antifungal antibacterial antibiotics were added to selectively inhibit the growth due to contamination. Thus the change in color and eventual growth of a colony give presumptive evidence of the presence of dermatophytes.

The diagnostic test RapidVet-D was developed using Taplin's work as an information base, but the components of the test were newly selected based on a new definition of the objective. The test was designed to be used directly on samples taken in veterinary practice from active lesions, simplifying the procedure and allowing a specific diagnosis of dermatophyte infection, or lack thereof, to be made in a few days while avoiding the common forms of external contamination. The underlying philosophy of this test is that, in most cases, for the practicing veterinarian, a rapid yes/no result with overall accuracy of 97.8% is preferable to obtaining results approaching 100% accuracy in 12-14 days and that microscopic identification is not necessary.

This report examines the specificity, sensitivity, and reproducibility in the field of RapidVet-D results on a statistically significant number of samples taken from different environments and subject to different kinds of contamination.

FIELD EVALUATION

A total of 1681 samples were used in the field, comprised of 820 samples from dogs, 369 from cats

and 492 from horses. The results obtained from this evaluation are shown in Table 1 below.

On the basis of these results, using color change after 72 hours as the only indication for the presence of dermatophytes, RapidVet-D demonstrated an accuracy rate of 97.7% (out of 1681 samples tested, 1643 samples were properly categorized). 510 samples exhibited a color change within 72 hours (a positive result) and 1171 samples did not exhibit a color change within 72 hours (a negative result).

After continued incubation for a total period of 14 days, followed by macroscopic and microscopic examination, 515 samples were confirmed positive and 1166 samples were confirmed negative. 17 of the samples exhibiting a color change during the 72 hour period proved negative for dermatophytes after 14 days and were thus considered to be false positives, resulting in a total false positive ratio of 3.3% of confirmed negative samples. 21 samples which did not exhibit a color change within 72 hours subsequently revealed the presence of dermatophytes, resulting in a false negative rate of 1.8% of confirmed positive samples.

It should be noted that the false negative samples mainly resulted not because of a lack of eventual color change of the culture medium, but only because of the fact that the color change was observed after a period of 72 hours, the period fixed for verifying the positivity/negativity of the samples. The delay in the color change may have been due to one of the following reasons:

- reaction tube not brought to room temperature (22-25°C or 72-77°F) immediately before and during the entire period of use
- sample not taken correctly from area of the lesion
- incorrect placement of the sample on the reaction substrate
- presence of slow metabolizing strains of dermatophytes

It should also be noted that the method of taking the sample was very important - the area should be gently cleaned with sterile gauze or 70% alcohol, which removes all traces of potential contamination, and the sample taken from the area surrounding the lesion.

The dermatophytes most commonly found were:

Microsporum canis
Trichophyton equinum
Microsporum gypseum
Trichophyton mentagrophytes

In **dogs**, 68.6% of positives were caused by *M. canis*, 17.8% by *M. gypseum*, 11.6% by *T. mentagrophytes* and 2.0% by *T. rubrum*. In **cats**, 88.8% of positives were caused by *M. canis*, 4.5% by *M. gypseum* and 6.7% by *T. mentagrophytes*. In **horses**, 72.5% were caused by *T. equinum*, 11.5% by *M. canis*, 12.0% by *M. gypseum* and the remaining 4.0% by *T. mentagrophytes*.

CONCLUSIONS

Based on these results, it is possible to conclude that RapidVet-D represents a valid and rapid method (24-72 hours) for screening for the presence of dermatophyte infections in dogs, cats and horses. This study's evaluation of RapidVet-D demonstrates the statistically high level of sensitivity, specificity and reproducibility of this test, even when color change within 72 hours is used as the only element for a positive conclusion.

When used within the parameters of the test's intended design, (i.e., that, in most cases, for the practicing veterinarian, a rapid test result with low incidence of false positive and false negative results is preferable to waiting 12-14 days for results with greater accuracy, and that microscopic identification is not necessary), RapidVet-D can be a useful and convenient addition to the veterinary practitioner's clinical laboratory.

Table I. Results of Field Evaluation

	Canine	Feline	Equine
Total samples evaluated	820	369	492
Total positive samples by color change	282 (34.39%)	85 (23.04%)	143 (29.07%)
Total negative samples by color change	538 (65.61%)	284 (76.96%)	349 (70.93%)
Total false positives (% of confirmed negatives)	9 (1.68%)	3 (1.06%)	5 (1.44%)
Total false negatives (% of confirmed positives)	11 (3.87%)	4 (4.65%)	6 (4.17%)
Accuracy	97.55%	98.10%	97.77%

RapidVet-D is manufactured in Italy by Agrolabo S.p.A.
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