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## ANIMAL HUSBANDRY

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### THE STUDY OF THE SENSITIZING PROPERTIES OF MYCOBACTERIA OF VARIOUS SPECIES

Research article

#### Abstract

The purpose of the study: selection of dosages of tuberculins to control the intensity of the development of delayed-type hypersensitivity in guinea pigs sensitized by various types of mycobacteria. Studies have shown that it is advisable to evaluate the sensitizing properties of non-tuberculous mycobacteria and avian mycobacteria at the same time using different doses of PPD for mammals and PPD for birds. To assess the delayed-type hypersensitivity state in guinea pigs sensitized by bovine mycobacteria, non-tuberculous mycobacteria and avian mycobacteria, optimal doses of PPD for birds are: 25 and 5 IU, and PPD for mammals: 25 and 5 IU.

**Keywords:** mycobacteria, dose of mycobacteria, guinea pigs, sensitization, delayed-type hypersensitivity, PPD for mammals, PPD for birds

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### ИЗУЧЕНИЕ СЕНСИБИЛИЗИРУЮЩИХ СВОЙСТВ МИКОБАКТЕРИЙ РАЗЛИЧНЫХ ВИДОВ

Научная статья

#### Аннотация

Цель исследования: подбор дозировок туберкулинов для контроля интенсивности развития гиперчувствительности замедленного типа у морских свинок сенсibilизированных разными видами микобактерий. Проведенные исследования продемонстрировали, что оценку сенсibilизующих свойств нетуберкулезных микобактерий и микобактерий птичьего вида целесообразно проводить одновременно используя различные дозировки ППД для млекопитающих и ППД для птиц.

Для оценки состояния ГЗТ у морских свинок сенсibilизированных микобактериями бычьего вида, нетуберкулезными микобактериями и микобактерий птичьего вида оптимальными дозами ППД для птиц являются: 25 и 5 МЕ, а ППД для млекопитающих: 25 и 5 МЕ.

**Ключевые слова:** микобактерии, доза микобактерий, морские свинки, сенсibilизация, гиперчувствительность замедленного типа, ППД для млекопитающих, ППД для птиц.

#### 1. Introduction

More than a hundred years ago, Robert Koch described the property of mycobacteria to cause a delayed-type hypersensitivity state (DTH) when administered to guinea pigs [1].

The ability of mycobacteria to cause DTH in the animal body is used in the biological assessment of different types of mycobacteria. One of the tests of biological assessment of mycobacteria is the study of sensitizing properties.

The sensitizing properties of bovine and human type mycobacteria in guinea pigs are carried out using tenths of a milligram of mycobacteria.

The sensitizing properties of avian mycobacteria and atypical mycobacteria are evaluated using milligrams or more [2], [3].

It is known that the introduction of high doses of mycobacteria into the body of animals is accompanied by immunosuppression [4].

Therefore, to obtain representative results of a study of sensitizing properties, it is necessary to use minimum dosages of avian mycobacteria and atypical mycobacteria.

The sensitizing properties of mycobacteria can be evaluated using various dosages of tuberculin.

In addition, most authors evaluate the sensitizing properties of mycobacteria using one dose of PPD for mammals or one dose of PPD for birds [4].

Using this approach does not allow to evaluate the sensitizing properties of different types of non-tuberculous mycobacteria with different sensitizing ability.

Given the foregoing objective of the study was: the selection of dosages of tuberculin to control the intensity of development of DTH in guinea pigs sensitized by different types of mycobacteria.

## 2. Material and methods

The study used female albino guinea pigs weighing 500-650 grams. A total of 30 guinea pigs were used in the studies, distributed in five groups of six animals each.

The following types of mycobacteria were used in the work:

*M. bovis* strain BCG;

*M. avium* strain 2282 (Ranyon group III);

*M. intracellulare* strain S-13 C (Ranyon group III);

*M. scrofulaceum* strain N-12 C (Ranyon group II);

*M. fortuitum* strain 18023 (Ranyon group IV);

Mycobacteria were grown on Pavlovsky's medium for 30 days at a temperature of 37 ° C.

From bird mycobacteria and atypical mycobacteria, suspension variants were prepared in physiological saline containing 1 mg of mycobacteria in a dose.

From bovine mycobacteria, a suspension was prepared in physiological saline with a dose of 0.2 mg / 0.1 cm<sup>3</sup>.

The study used tuberculins (PPD for mammals; PPD for birds) produced by the Kursk Biofactory, standardized by biological activity relative to International Standards (PPD bovine and PPD avium).

Dilutions of tuberculins were carried out on phosphate buffer (pH 7.0).

Statistical processing of the experimental results was carried out using Microsoft office Excel software.

## 3. Result and discussion

30 days after sensitization of guinea pigs intracutaneously, dilutions of PPD for mammals were introduced: 25 IU / 0.1 cm<sup>3</sup>, 5 IU / 0.1 cm<sup>3</sup>, 1 IU / 0.1 cm<sup>3</sup> and dilutions of PPD for birds: 125 IU / 0.1 cm<sup>3</sup>, 25 IU / 0.1 cm<sup>3</sup>, 5 / 0.1 cm<sup>3</sup> on the right and left, respectively.

The use of dosages of PPD for mammals (25 IU, 5 IU, 1 IU) and PPD for birds (125 IU, 25 IU, 5 IU) is due to the linear dose-response DTH. The criteria for evaluating the used doses of PPD for mammals and PPD for birds were: the intensity of the reaction and the percentage of the reaction to the used dose in the group of guinea pigs. The results of the study are presented in the table.

Table 1 – The use of different doses of PPD for mammals and PPD for birds in the study of the sensitizing properties of mycobacteria

Options	Sensitization <i>M. bovis</i> strain BCG, 1 group of guinea pigs					
	PPD for birds			PPD for mammals		
	125 IU	25 IU	5 IU	25 IU	5 IU	1 IU
M±m	15,58±0,83	11,33±0,83	0±0	19,67±0,86	15,83±0,82	13,08±0,87
%	83	33	0	100	100	100
	Sensitization <i>M. avium</i> strain 2282, 2 group of guinea pigs					
M±m	20,08±0,66	16,08±1,06	13,42±0,81	13,58±1,04	0±0	0±0
%	100	100	100	100	0	0
	Sensitization <i>M. intracellulare</i> strain S-13 C, 3 group of guinea pigs					
M±m	12,58±0,71	7,75±1,67	2,67±1,71	0±0	0±0	0±0
%	100	83	33	0	0	0
	Sensitization <i>M. scrofulaceum</i> strain N-12 C, 4 group of guinea pigs					
M±m	15,75± 0,73	11,67± 0,36	4,42± 2,01	8,0± 2,57	1,67	0± 0
%	100	100	50	67	17	0
	Sensitization <i>M. Fortuitum</i> strain 18023, 5 group of guinea pigs					
M±m	0	0	0	0	0	0
%	0	0	0	0	0	0

From the data obtained, it can be seen that in the group of guinea pigs sensitized with *M. bovis*, the use of PPD for birds at doses of 125 and 25 IU makes it possible to reveal the state of DTH in 50% and 33%, respectively, while using a dose of 5 IU does not allow to detect DTH. The use of PPD for mammals in a dose of 25 IU, 5 IU, and 1 IU in the first group of guinea pigs makes it possible to characterize the state of DTH in 100%.

Thus, when evaluating the sensitizing properties of bovine mycobacteria, it is advisable to use PPD for birds at doses of 25 and 5 IU, and PPD for mammals at doses of 25 and 5 IU.

In the second group of guinea pigs sensitized by avian mycobacteria, it can be seen that the PPD for birds in doses of 125 IU, 25 IU, 5 IU makes it possible to detect DTH in 100%. At the same time, the use of PPD for mammals makes it possible to detect DTH in 100% only when using a dose of 25 IU. DTH response to doses of 5 IU and 1 IU PPD for mammals in the guinea pig group of sensitized *M. avium* was not detected.

Thus, when evaluating the sensitizing properties of avian mycobacteria, it is advisable to use PPD for birds at doses of 25 and 5 IU, and PPD for mammals at doses of 25 and 5 IU.

In the third group of guinea pigs sensitized with *M. intracellulare*, it is seen that PPD for birds at a dose of 125 IU allows us to identify the state of DTH in 100%, at a dose of 25 IU in 83%, at a dose of 5 IU - 33%. The use of different doses of PPD for mammals: 25 IU, 5 IU, 1 IU does not reveal the state of DTH.

Thus, when assessing the sensitizing properties of *M. intracellulare* mycobacteria, it is advisable to use PPD for birds at doses of 25 and 5 IU, and PPD for mammals at doses of 25 and 5 IU.

In the fourth group of guinea pigs sensitized with *M. scrofulaceum*, it is seen that the PPD for birds in doses of 125 IU and 25 IU makes it possible to detect DTH in 100%. The use of a dose of 5 IU PPD for birds reveals the state of DTH in only 50%. Using a dose of 25 IU PPD for mammals reveals DTH in 67%. The use of a dose of 5 IU PPD for birds reveals the state of DTH in only 50%. Using a dose of 25 IU PPD for mammals reveals DTH in 67%. Using a dose of PPD for birds of 5 IU can detect DTH in 17%. The use of a dose of PPD for mammals of 1 IU does not reveal the state of DTH.

Thus, when assessing the sensitizing properties of *M. scrofulaceum* mycobacteria, it is advisable to use PPD for birds at doses of 25 and 5 IU, and PPD for mammals at doses of 25 and 5 IU.

At the same time, it was found that the use of different dosages of PPD for mammals and PPD for birds does not allow us to evaluate the sensitizing properties of atypical mycobacteria *M. fortuitum*, which requires additional studies.

Studies have shown the possibility of sharing different doses of PPD for mammals and PPD for birds in assessing the sensitizing properties of mycobacteria of various species.

#### 4. Conclusion

1. Studies have shown that it is advisable to evaluate the sensitizing properties of non-tuberculous mycobacteria and avian mycobacteria at the same time using different doses of PPD for mammals and PPD for birds.

2. To assess the delayed-type hypersensitivity state in guinea pigs sensitized by bovine mycobacteria, non-tuberculous mycobacteria and avian mycobacteria, optimal doses of PPD for birds are: 25 and 5 IU, and PPD for mammals: 25 and 5 IU.

#### Conflict of Interest

None declared.

#### Конфликт интересов

Не указан.

#### References

1. Безгин В.М. Основы промышленной иммунобиотехнологии / В.М. Безгин, Н.Н. Быкова, В.Е. Козлов, А.А. Нежуга, А.В. Сверчков. – Курск.: КГСХА, 2011. – 511 с.
2. Мясоедов Ю.М. Оценка методов контроля качества микобактериальных аллергенов изготавливаемых с использованием *M. bovis*// Вестник КГСХА. – 2015. – №8. – С. 209-212.
3. Мясоедов Ю.М. Оценка методов контроля качества аллергенов микобактерий *M. avium-intracellulare* и *M. scrofulaceum*// Вестник КГСХА. – 2017.-№6. – С.19-25.
4. Найманов А.Х. Микобактериальные инфекции крупного рогатого скота (туберкулёз, паратуберкулёз) / А.Х. Найманов, М.И. Гулюкин. – М.: «Зооветкнига», 2014. – 235 с.

#### References in English

1. Bezgin V.M. Osnovi promichlennoi immunobiotehnologii [Fundamentals of Industrial Immunobiotechnology] / V. M. Bezgin, N. N. Bikova, V.E. Kozlov, A.A. Neguta, A.V. Sverchkov. – Kursk.:KGSXA, 2011. – 511 p. [in Russian]
2. Myasoedov Y. M. Ocenka metodov kontrolajj kachestva mikobakterialnich allergenov isgotavlivaemich s ispolsovaniem *M. bovis* [Evaluation of quality control methods for mycobacterial allergens manufactured using *M. bovis*] // Vestnik KGSXA, – 2015. – №8. – P.209-212. [in Russian]
3. Myasoedov Y. M. Ocenka metodov kontrolajj kachestva allergenov mikobakterii *M. avium-intracellulare* i *M. scrofulaceum* [Evaluation of quality control methods for allergens of mycobacteria *M. avium-intracellulare* and *M. scrofulaceum*] // Vestnik KGSXA, – 2017. – №6. – P.19-25. [in Russian]
4. Naimanov A.H. Micabakterialnie infectii krupnogo rogatogo skota (tuberculosis, paratuberculosis) [Mycobacterial infections of cattle (tuberculosis, paratuberculosis)] / A.H. Naimanov, M.I. Gulukin. – M.: Zoovetkniga, 2014. – 235 p. [in Russian]