Abraxis Tylosin Plate Kit

PN 52256B

Instructional Booklet
Read Completely Before Use.

INTENDED USE

The Tylosin Plate Kit is a competitive ELISA for the quantitative analysis of Tylosin in honey products.

ASSAY PRINCIPLES

The Tylosin plate kit is a competitive enzyme-labeled immunoassay. Tylosin is extracted from a sample by blending or shaking with extraction solution. The Tylosin sample extract and calibrators are pipetted into the test wells followed by Tylosin antibody into the test wells to initiate the reaction. During the 30 minute incubation period, Tylosin from the sample and Tylosin HRP conjugate compete for binding to Tylosin antibody. Following this 30 minute incubation, the contents of the well are removed and the wells are washed to remove any unbound Tylosin, Tylosin HRP conjugate and free Tylosin antibody. After wash with wash solution, a clear substrate is then added to the wells and any bound enzyme conjugate causes the conversion to a blue color. Following a 30 minute incubation, the reaction is stopped and the amount of color in each well is read. The color of the unknown samples is compared to the color of the calibrators and the Tylosin concentration of the samples is derived.

SPECIFICITY:

The Tylosin Plate Kit can not differentiate between the various Tylosins, but detects their presence to differing degrees. The following table shows the % cross reactivity of Tilmicosin versus Tylosin. All concentrations are in parts per billion (ppb).

Compound	% CR
Tylosin	**100%**
Tilmicosin	**125%**

DETECTION LIMIT:

Honey: 1.25 ppb

REAGENTS AND MATERIALS PROVIDED

The kit in its original packaging can be used until the end of the month indicated on the box label when stored at 2 – 8ºC.

- Plate containing 12 test strips of 8 wells each vacuum-packed in aluminized pouch with indicating dessicant.
- 6 vials each containing 2 mL of Tylosin calibrators corresponding to 0, 0.05, 0.1, 0.5, 1, 5 µg/L (ppb) of Tylosin.
- 1 vial containing 7 mL Tylosin HRP Enzyme Conjugate.
- 1 vial containing 7 mL of anti-Tylosin antibody.
- 1 vial containing 14 mL of Substrate.
- 1 vial containing 14 mL of Stop Solution. (Caution! 1N HCl. Handle with care.)
- Instructions

PRECAUTIONS

1. Each reagent is optimized for use in the Tylosin Plate Kit. Do not substitute reagents from any other manufacturer into the test kit. Do not combine reagents from other Abraxis Tylosin Plate Kits with different Lot numbers.

2. Dilution or adulteration of reagents or samples not called for in the procedure may result in inaccurate results.

3. Do not use reagents after expiration date.

4. Reagents should be brought to room temperature, 20 – 28ºC (62 – 82ºF) prior to use. Avoid prolonged (> 24 hours) storage at room temperature.

5. Tylosin is an antibiotic and should be treated with care.

6. The Stop Solution is 1N hydrochloric acid. Avoid contact with skin and mucous membranes. Immediately clean up any spills and wash area with copious amounts of water. If contact should occur, immediately flush with copious amounts of water.

MATERIALS REQUIRED BUT NOT PROVIDED

1. Laboratory quality distilled or deionized water.
2. Graduated cylinder, 100 ml or larger.
3. Glassware for sample extraction and extract collection.
4. Methanol
5. Pipet with disposable tips capable of dispensing 50 µL.
6. Multi-channel pipet; 8 channel capable of dispensing 50 and 100 µL.
7. Paper towels or equivalent absorbent material.
8. Microwell plate or strip reader with 450nm filter.
9. Timer
10. Vortex mixer
11. Wash bottle
12. 20 mM PBS, pH 7.4. [0.62 g NaH_2PO_4-$2H_2O$+ 5.73 g Na_2HPO_4-$12H_2O$ + 9 g NaCl, /liter, pH 7.4]

HONEY SAMPLE EXTRACTION (1:25 dilution)

1. Weigh 1 gram of honey in a screw cap glass bottle (50ml size)
2. Add 24 ml of 20 mM PBS (1ml sample + 24 ml buffer, 1:50 dilution)
3. Put the sample bottle in an ultrasonic water bath for 5 min.
4. Mix vigorously for 2 min.
5. Before transfering 50 ul for the assay, invert the sample bottle several times to mix.

TEST PROCEDURE (Note: Running calibrators and samples in duplicate will improve assay precision and accuracy.)

1. Allow reagents and sample extracts reach room temperature prior to running the test. Fill a wash bottle with lab grade water.

2. Place the appropriate number of test wells and into a microwell holder. Be sure to re-seal unused wells in the zip-lock bag with dessicant.

3. Using a pipet with disposable tips, add **50 µL enzyme conjugate** to the appropriate test wells. Be sure to use a clean pipet tip for each. Add **50 ul of Calibrators or Sample extract** to each well

4. Dispense **50 µL of Antibody Solution** into each test well.

5. Shake the plate gently for 30 seconds and incubate the test wells for **60 minutes**.

7. Dump the contents of the wells into an appropriate waste container. Fill the wells to overflowing with tap water and dump. Repeat 3X for a total of four washes.

8. Following the last wash tap the inverted wells onto absorbent paper to remove the last of the wash solution.

9. Dispense **100 µL of Substrate** into each well.

10. Incubate the wells for **30 minutes**.

11. Dispense **100 µL of Stop Solution** into each test well.

12. Read and record the absorbance of the wells at 450nm using a strip or plate reader.

RESULTS INTERPRETATION

1. Semi-quantitative results can be derived by simple comparison of the sample absorbances to the absorbance of the calibrator wells: Sample containing less color than a calibrator well having a concentration of Tylosin greater than the concentration of the calibrator. Samples containing more color than a calibrator well have a concentration less than the concentration of the calibrator.

2. Quantitative interpretation requires graphing the absorbances of the calibrators (X axis) versus the log of the calibrator concentration (Y axis) on semi-log graph paper. A straight line is drawn through the calibrator points and the sample absorbances are located on the line. The corresponding point on the Y axis is the concentration of the sample. Samples with absorbances greater than the lowest calibrator or less than the highest calibrator must be reported as < 0.05 ppb or >5 ppb, respectively.

   Alternatively, Abraxis can supply a spreadsheet template which can be used for data reduction. Please contact Abraxis for further details.

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GENERAL LIMITED WARRANTY

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In the event of a breach of the foregoing warranty, Abraxis's sole obligation shall be to repair or replace, at its option, any product or part thereof that proves defective in materials or workmanship within the warranty period, provided the customer notifies Abraxis promptly of any such defect. The exclusive remedy provided herein shall not be deemed to have failed of its essential purpose so long as Abraxis is willing and able to repair or replace any nonconforming Abraxis product or part. Abraxis shall not be liable for consequential, incidental, special or any other indirect damages resulting from economic loss or property damage sustained by a customer from the use of its products. However, in some states the purchaser may have rights under state law in addition to those provided by this warranty.

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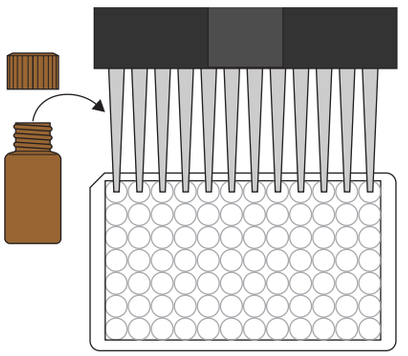
Tylosin Plate, Detailed ELISA Procedure

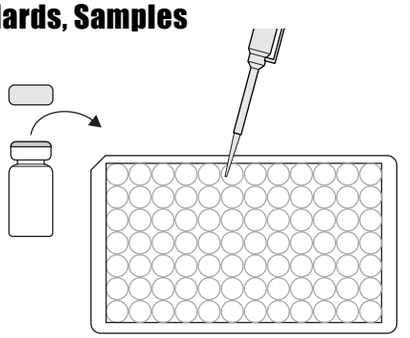
1. Addition of Enzyme Conjugate

Add 50 ul of enzyme conjugate to the wells of the test strips successively using a multi-channel pipette or a stepping pipette according to the working scheme given. We recommend using duplicates or triplicates.



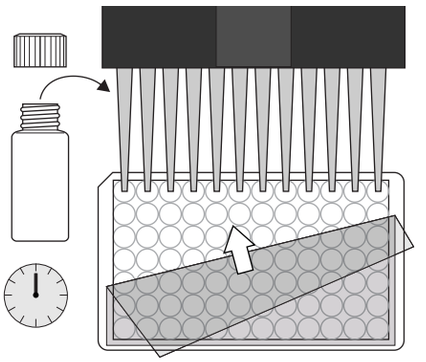
2. Addition of Standards, Samples

Add 50 ul of the standard solutions or samples to the wells of the test strips according to the working scheme given.



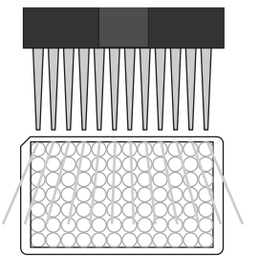
3. Addition of Antibody Solution

Add 50 uL of the antibody solution to the individual wells successively using a multi- channel pipette or a stepping pipette. Cover the wells with parafilm or tape and mix the contents by moving the strip holder in a rapid circular motion on the benchtop. Be careful not to spill contents. Incubate the strips for 60 min at room temperature.

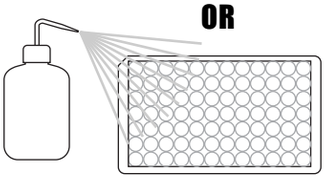


4. Washing of Plates

After incubation, remove the covering and vigorously shake the contents of the wells into a sink. Wash the strips four times with a multi-channel pipette or wash bottle using the diluted 1X washing buffer solution. Please use at least a volume of 250 uL of washing buffer for each well and each washing step. Remaining buffer in the wells should be removed by patting the plate dry on a stack of paper towels.

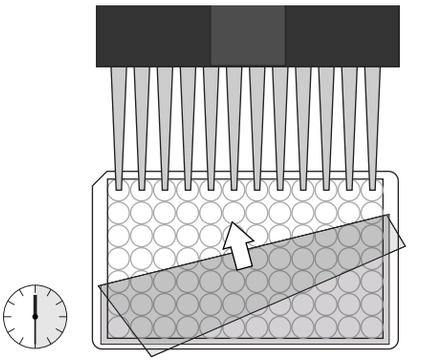


OR



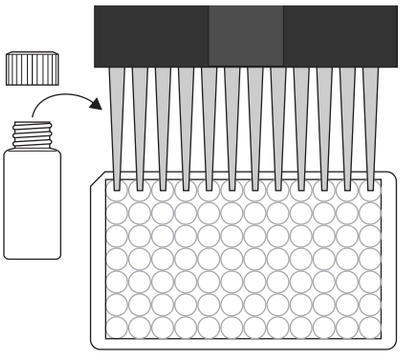
5. Addition of Substrate/Color Solution

Add 100 uL of substrate/color solution to the individual wells successively using a multi-channel pipette or a stepping pipette. Cover the wells with parafilm or tape and mix the contents by moving the strip holder in a rapid circular motion on the benchtop. Be careful not to spill contents. Incubate the strips for 30 min at room temperature.



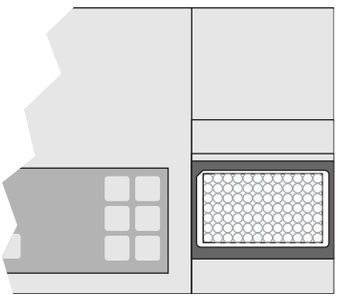
6. Addition of Stopping Solution

Add 100 uL of stop solution to the wells in the same sequence as for the substrate solution using a multi- channel pipette or a stepping pipette.



7. Measurement of Color

Read the absorbance at 450 nm using a microplate ELISA reader. Calculate results.



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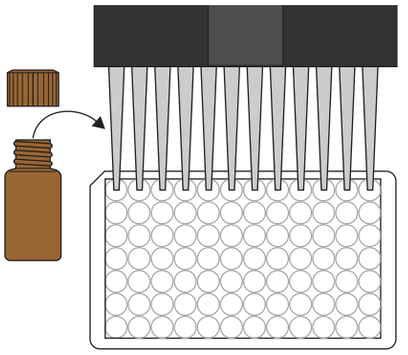
Tylosin Plate, Concise ELISA Procedure

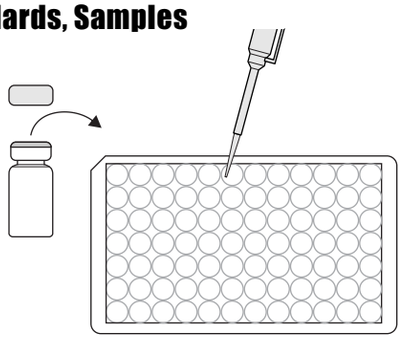
1. Addition of Enzyme Conjugate

Add 50 ul of enzyme conjugate.



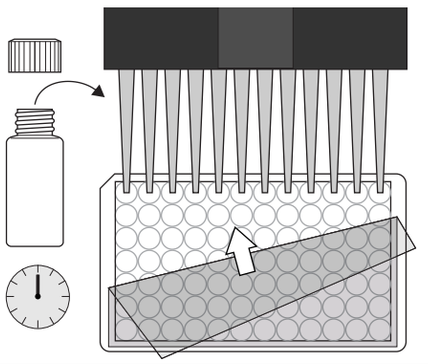
2. Addition of Standards, Samples

Add 50 ul of the standard solutions or samples.



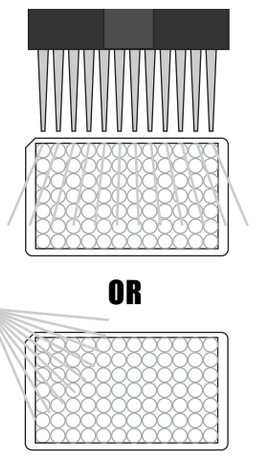
3. Addition of Antibody Solution

Add 50 uL of the antibody solution. Cover and mix for 30 seconds by rotating on benchtop. Incubate for 60 min at room temperature.



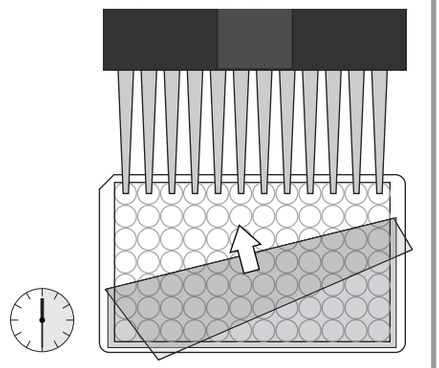
4. Washing of Plates

Wash the plates four times with 250 uL of diluted 1X washing buffer.



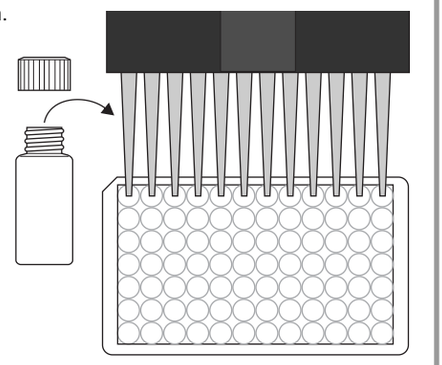
5. Addition of Substrate/Color Solution

Add 100 uL of substrate/color solution. Incubate 30 minutes at room temperature and away from direct sunlight.



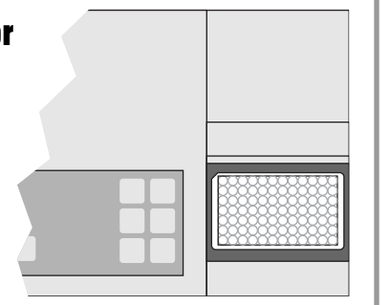
6. Addition of Stopping Solution

Add 100 uL of stop solution.



7. Measurement of Color

Measure color at 450 nm. Calculate results.



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Safety Data Sheet

Section 1: Product and Company Identification

1.1 Product Identifiers:
Product Name: Tylosin ELISA Plate Kit
Product Code: 52256B
1.2 Identified Use: Determination of Tylosin in samples. **Restrictions on Use:** For research use only.
1.3 Company: Abraxis, Inc.**,** 124 Railroad Drive**,** Warminster, PA 18974 USA**,** info@abraxiskits.com +1(215) 357-3911, FAX +1(215) 357-5232
1.4 Emergency Telephone Number: +1(215) 357-3911

Section 2: Hazard(s) Identification

2.1 Classification of the mixture: Not a hazardous mixture.
2.2 GHS Label elements, including precautionary statements: Not applicable.
2.3 Hazards not otherwise classified (HNOC) or not covered by GHS: None known.
2.4 Unknown acute toxicity: None known.

Section 3: Composition / Information on Ingredients

3.2 Mixtures: *Contains no hazardous ingredients at levels requiring disclosure by the OSHA Hazard Communication Standard (29 CFR 1910.1200)*, however it contains minor amounts of materials considered hazardous. We recommend handling all substances with caution.

Section 4: First Aid Measures

4.1 Description of first aid measures: Consult a physician. Show this safety data sheet to the doctor in attendance. Move out of dangerous area.
If inhaled: If breathed in, move person into fresh air. If not breathing, give artificial respiration. Consult a physician.
In case of skin contact: Wash off with soap and plenty of water. Consult a physician.
In case of eye contact: Rinse thoroughly with plenty of water for at least 15 minutes and consult a physician.
If swallowed: Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician.
4.2 Most important symptoms and effects, both acute and delayed: No data available
4.3 Indication of any immediate medical attention and special treatment needed: No data available. Treat symptomatically.

Section 5: Fire-fighting Measures

5.1 Suitable extinguishing media: Use an extinguishing agent suitable for the surrounding fire.
5.2 Special hazards arising from the substance or mixture: None known
5.3 Advice for firefighters: Wear self-contained breathing apparatus for fire-fighting if necessary.
5.4 Further information: No data available

Section 6: Accidental Release Measures

6.1 Personal precautions, protective equipment and emergency procedures: Use personal protective equipment (see section 8). Avoid dust formation. Avoid breathing vapors, mist, dust, or gas. Ensure adequate ventilation. Evacuate personnel to safe areas.
6.2 Environmental precautions: Prevent further leakage or spillage if safe to do so. Do not let product enter drains. Discharge into the environment must be avoided.
6.3 Methods and materials for containment and cleaning up: Solids (if applicable): Pick up and arrange disposal without creating dust. Sweep up and shovel. Liquids (if applicable): Absorb with liquid-binding material (sand, diatomite, acid binders, universal binders, sawdust). Keep in suitable, closed containers for disposal.
6.4 Reference to other sections: For information on safe handling see section 7.
For information on personal protection see section 8.
For information on disposal see section 13.

Section 7: Handling and Storage

7.1 Precautions for safe handling: See section 2. Avoid inhalation of vapors and contact with skin and eyes. Wear appropriate personal protective equipment. Do not eat, drink, or smoke in work area.
7.2 Precautions for safe storage: Keep container(s) tightly closed in a dry, well-ventilated place. Protect from physical damage. See label or product insert for appropriate storage temperature and additional specific information.

7.3 Specific end use(s): No data available

Section 8: Exposure Controls / Personal Protection

8.1 Control parameters: Not applicable.
8.2 Exposure controls:
Appropriate engineering controls: Provide adequate ventilation. Handle in accordance with good industrial hygiene and safety practice. Wash hands before breaks and at the end of workday. Keep away from food and beverages.
Personal protective equipment: The usual precautionary measures, including eye/face/skin protection, should be taken when handling any chemical. Avoid contact with eyes, skin, and clothing.
Eye protection: As with handling of any chemical, wear approved safety goggles.
Skin protection: Handle with gloves. No specific information regarding glove material or thickness is available, but gloves must be impermeable and resistant to the substance being handled. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands.
Respiratory protection: As with any chemical, where excessive vapor, mist, or dust may result, use a chemical fume hood or approved respiratory protection equipment.
Body protection: Lightweight, protective clothing.

Section 9: Physical and Chemical Properties

9.1 Information on basic physical and chemical properties of the mixture
Appearance: Multiple
Odor: Characteristic
Odor Threshold: No data available
pH: Multiple
Melting point/freezing point: No data available
Initial boiling point and boiling range: No data available
Flash point: Not applicable
Evaporation rate: No data available
Flammability (solid, gas): No data available
Upper/lower flammability or explosive limits No data available
Vapor pressure: No data available
Vapor density: No data available
Relative density: No data available
Water solubility: Various
Partition coefficient: n-octanol/water: No data available
Auto-ignition temperature: Not applicable
Decomposition temperature: No data available
Viscosity: No data available
Explosive properties: No data available
Oxidizing properties: No data available
9.2 Other information: No data available

Section 10: Stability and Reactivity

10.1 Reactivity: No data available
10.2 Chemical stability: Stable under recommended storage conditions.
10.3 Possibility of hazardous reactions: No data available
10.4 Conditions to avoid: No data available
10.5 Incompatible materials: No data available
10.6 Hazardous decomposition products: No data available. In the event of fire: see section 5.

Section 11: Toxicological Information

11.1 Information on toxicological effects
Acute toxicity: Not available. To the best of our knowledge, the chemical, physical, and toxicological properties of this product have not been thoroughly investigated.
Inhalation: No data available **Ingestion:** No data available
Skin contact: Irritant to skin and mucous membranes.
Eye contact: May cause eye irritation in susceptible persons.

Respiratory or skin sensitization: No data available
Aspiration hazard: No data available
Mutagenicity: No data available
Carcinogenicity
IARC: No component of this product present at levels greater than or equal to 0.1% is identified as probable, possible or confirmed human carcinogen by IARC.
ACGIH: No component of this product present at levels greater than or equal to 0.1% is identified as a carcinogen or potential carcinogen by ACGIH.
NTP: No component of this product present at levels greater than or equal to 0.1% is identified as a known or anticipated carcinogen by NTP.
OSHA: No component of this product present at levels greater than or equal to 0.1% is identified as a carcinogen or potential carcinogen by OSHA.
Teratogenicity: No data available **Reproductive/fertility toxicity:** No data available
Specific target organ toxicity, single exposure: No data available
Specific target organ toxicity, repeated exposure: No data available

Section 12: Ecological Information

12.1 Toxicity: No data available
12.2 Persistence and degradability: No data available
12.3 Bioaccumulative potential: No data available
12.4 Mobility in soil: No data available
12.5 Results of PBT and vPvB assessment: No data available
12.6 Other adverse effects: An environmental hazard cannot be excluded in the event of unprofessional handling or disposal.

Section 13: Disposal Considerations

13.1 Waste treatment methods
Product: All waste must be handled and disposed according to local, state, and federal regulations. Avoid disposing large volumes in sewer.
Contaminated packaging: All waste must be handled and disposed according to local, state, and federal regulations.
Refer to sections 7 and 8 for safe handling guidance.

Section 14: Transport Information

UN Number: Not regulated **UN Proper shipping name:** Not classified as dangerous in the meaning of transport regulations.
Transport hazard class(es): No data available **Packing group:** No data available **Environmental hazard:** No data available
Bulk transport: No data available **Special considerations:** No data available

Section 15: Regulatory Information

To the best of our knowledge, this product contains no substances which, at their given concentrations, are considered hazardous by other regulatory agencies. Refer to section 3.

Section 16: Other information

This information is based on our present knowledge. While Abraxis , Inc. believes that the data contained herein are factual and the opinions expressed represent a best effort to present accurate information, the data are not to be taken as a warranty or representation for which Abraxis , Inc. assumes legal responsibility. The information shall not be taken as being all-inclusive and is to be used only as a guide. The data are offered solely for the user's consideration, investigation, and verification. These suggestions should not be confused with either state, municipal, or insurance requirements, or with national safety codes and constitute no warranty. Any use of these data and information must be determined by the user to be in accordance with applicable federal, state, and local regulations.

All materials and mixtures may present unknown hazards and should be used with caution. Since Abraxis , Inc. cannot control the methods, volumes, or conditions of use of this product, Abraxis , Inc. shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. An individual technically qualified to handle potentially hazardous chemicals must supervise the use of this material. This product is sold for research use only. It is not for any human or animal therapeutic or clinical diagnostic use.

Date this SDS was prepared: 5/24/2016
Version: 2
Changes from previous version: Abraxis, LLC changed to Abraxis, Inc.