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### **Near-IR imaging of FPI-749 conjugated PLGA microparticles**

**Purpose:** To evaluate the effectiveness of FPI-749 for labeling and rendering fluorescent polymer systems for use in visualization during in-vivo research.

#### **Methods:**

Amine endcapped PLGA-50:50 with a molecular weight of roughly 28kDa (PolyVivo intermediate product # AI10) was dissolved in anhydrous dichloromethane with stirring. To this stirring mixture the dry powdered Flamma Fluor dye FPI-749 was added directly and allowed to stir in dichloromethane overnight at room temperature. The resultant product was filtered and precipitated into cold hexane to purify. This polymer was processed into microparticles using a proprietary template based technique, sterilized by 70% ethanol and UV light exposure and freeze dried. Immediately prior to injection the microparticles were reconstituted in 0.9% sterile saline and injected subcutaneously into freshly shaved, female balb/c mice anesthetized with isoflurane. The injection site was marked with an indelible marker by a series of dots placed circularly around the site.

Under anesthesia, the mice were imaged using a 650nm class IIIa laser for excitation and a custom-modified commercial camera for emission collection. Imaging was done in a lab in the open with the lights on with no restrictions on access to the mice.

#### **Results**

Figures 1 and 2 below show the resultant images collected under standard brightfield imaging and near-IR modes. The brightfield image has the right ear and injection site markings labeled for orientation. In the near-IR mode the bright cluster inside the injection markings is the fluorescently labeled polymer indicating its successful visualization under the skin.

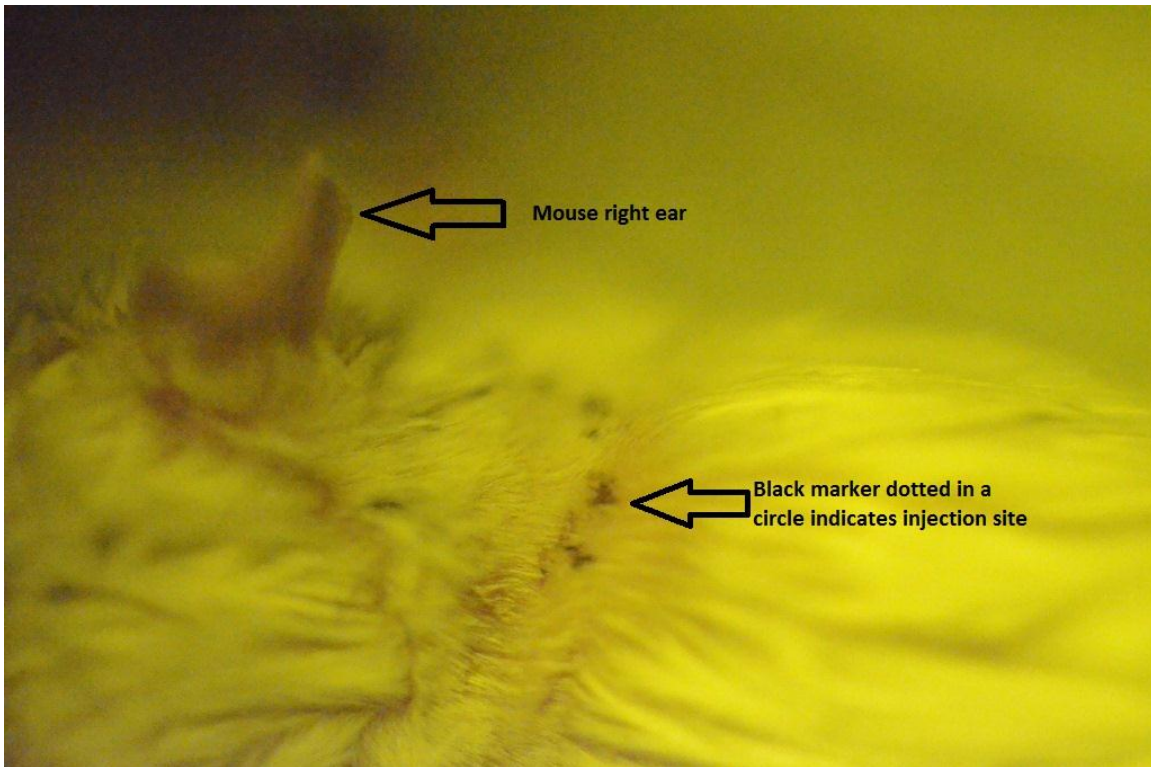


Figure 1. labeled brightfield image of mouse at injection site



Figure 2. Near-IR image showing fluorescently stained polymer visualized at injection site.

### Conclusion

Flamma Fluor FPI-749 can successfully react with amine endcapped polymers and the resultant product can be visualized in-vivo by near-IR imaging.