Supporting information

Carrier Gas Triggered Controlled Biolistic Delivery of DNA and Protein Therapeutics from Metal-Organic Frameworks

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MOF-Gun Components in Detail

Gas is delivered to the device from either a gas canister (Maddog Heavy Duty Paintball Tank Remote Coil connected to a tank, as shown) or 12 g CO_2 adapter (AMC Quick Charge 12 g CO_2 Adapter—not shown), either of which is attached to the Universal Fill Adapter (Ninja Paintball Universal Fill Adapter). Gas is from food-grade 12 g CO_2 cartridges or compressed air (oil-free membrane compressor with built-in water-oil filtration).

Gas enters by opening the needle valve, which pressurizes the system. A large analog pressure gauge (reading both SI and Imperial Units) indicates pressure. Once the desired pressure is reached, the needle valve is closed. The firing components of the MOF-Gun are composed of the following parts, assembled per Scheme S1. A high-pressure solenoid (Redhat Mfr #826H200) suitable for corrosive and inert liquid/gas applications with a maximum operating pressure of 3500 kPa is controlled via an adjustable relay (Dayton Time Delay Relay Mfr#6A855) interfaced to a Square D Relay Socket (Mfr #8501NR61) standard, octal, 11 pin configuration. A connected miniature firing push button activates the relay/solenoid.

Metal pipes and connectors are stainless steel rated at or above 3500 kPa and purchased in the United States from McMASTER-CARR. All components are designed and measured to imperial units (US standards). Thread and thread sizes of ¼ inch NPT. The tip is a prefabricated Solid-Stream Spray Nozzle manufactured by McMASTER CARR (Mfr# 7611T53). Washer inserts approximately have 0.325 cm inner diameter and 0.685 cm outer diameter and fit precisely within the tip.

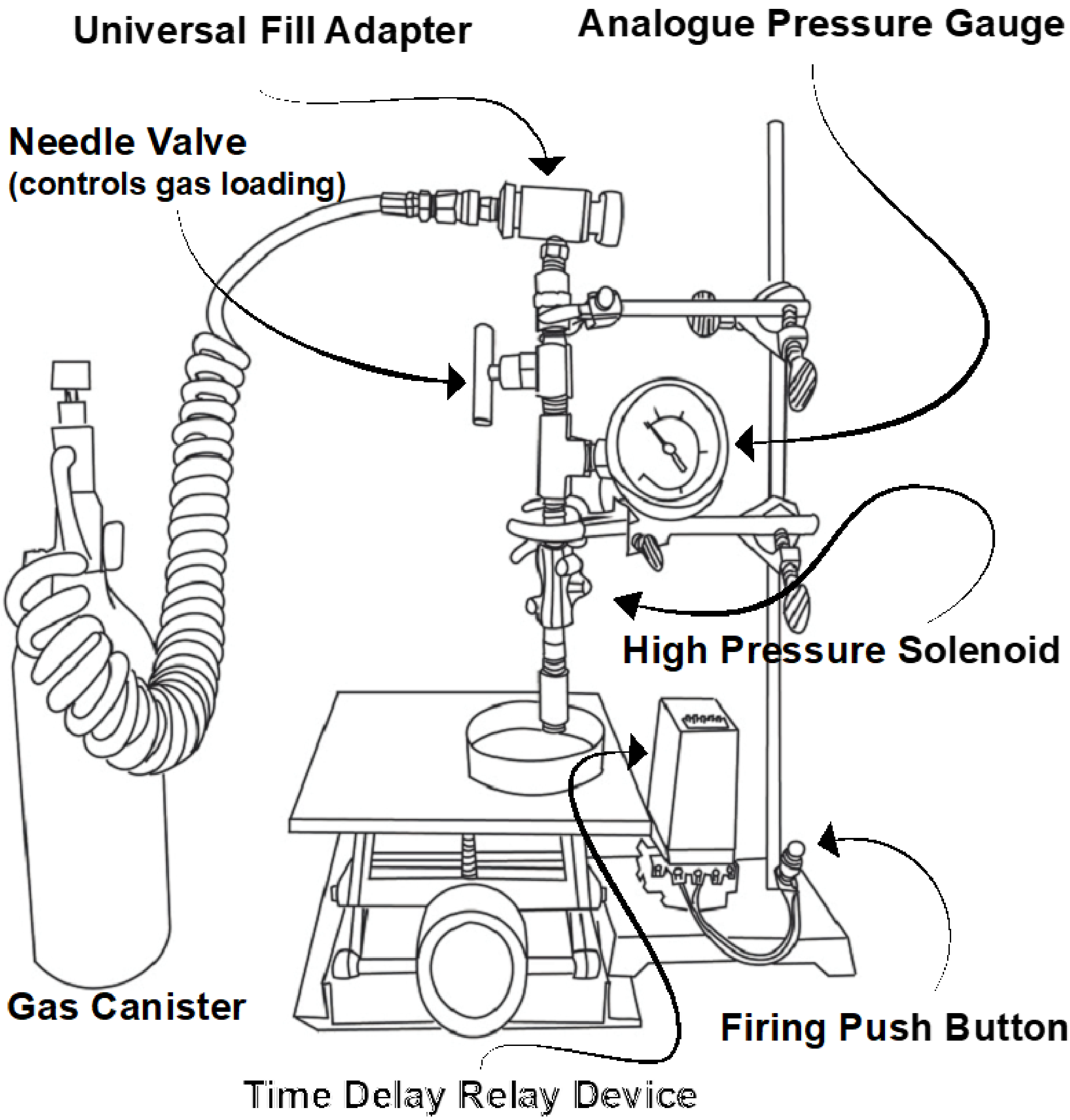
Loading and firing procedures:

A small piece of parafilm is sandwiched between two washers and then stretched to form a thin membrane to make the loading washers. The loading washers were then lodged into the gun nozzle. Then, 2.50 mg of powdered/solid sample of DsR@ZIF (5 µg of DsRed) was weighed and loaded onto the parafilm membrane. After that, the gun nozzle was screwed back into the MOF-Gun, and pressure and firing distance were adjusted. The relay timer was set to 0.9 s, and the firing button was pushed, releasing the particles. **CAUTION**: This can eject ZIF-8 at speeds sufficient to penetrate the skin. These experiments should be conducted with safety glasses, gloves, and lab coats. Discharging ZIF-Gun in a fume cabinet with airflow is recommended.

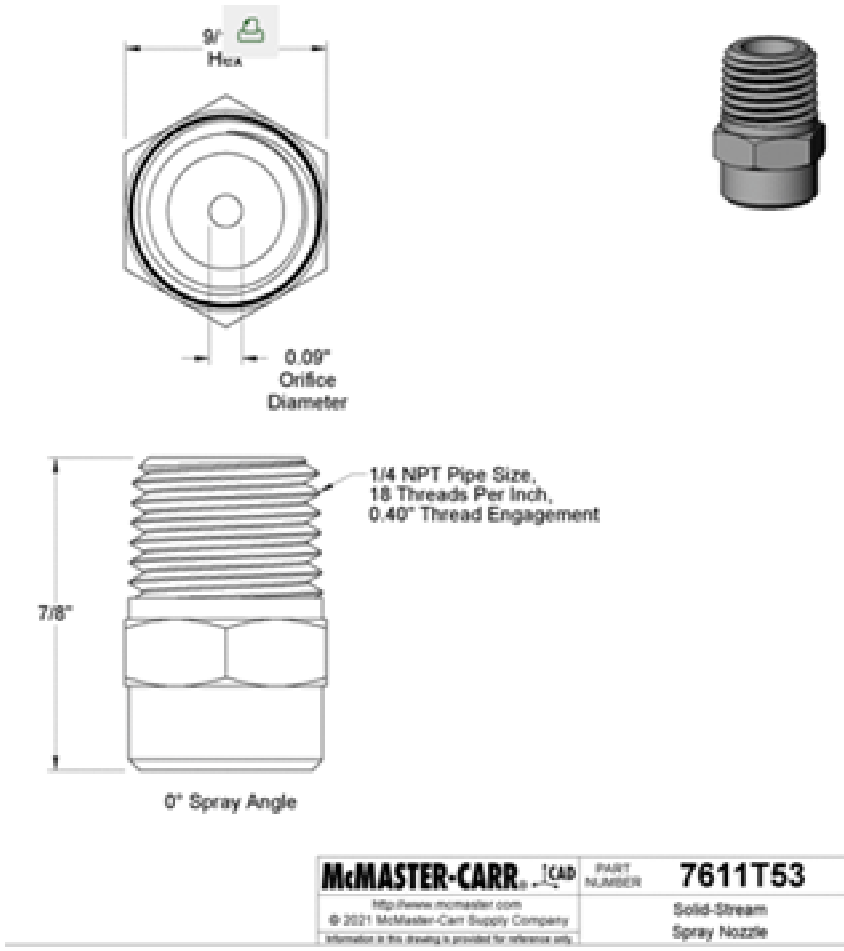
Time-resolved pH Meter

A time-resolved pH meter was constructed using a pH/ORP adapter (Phidgets.com) connected to a Phidget VINT Hub. The Hub interfaces to a PC. The following script was used to generate data:

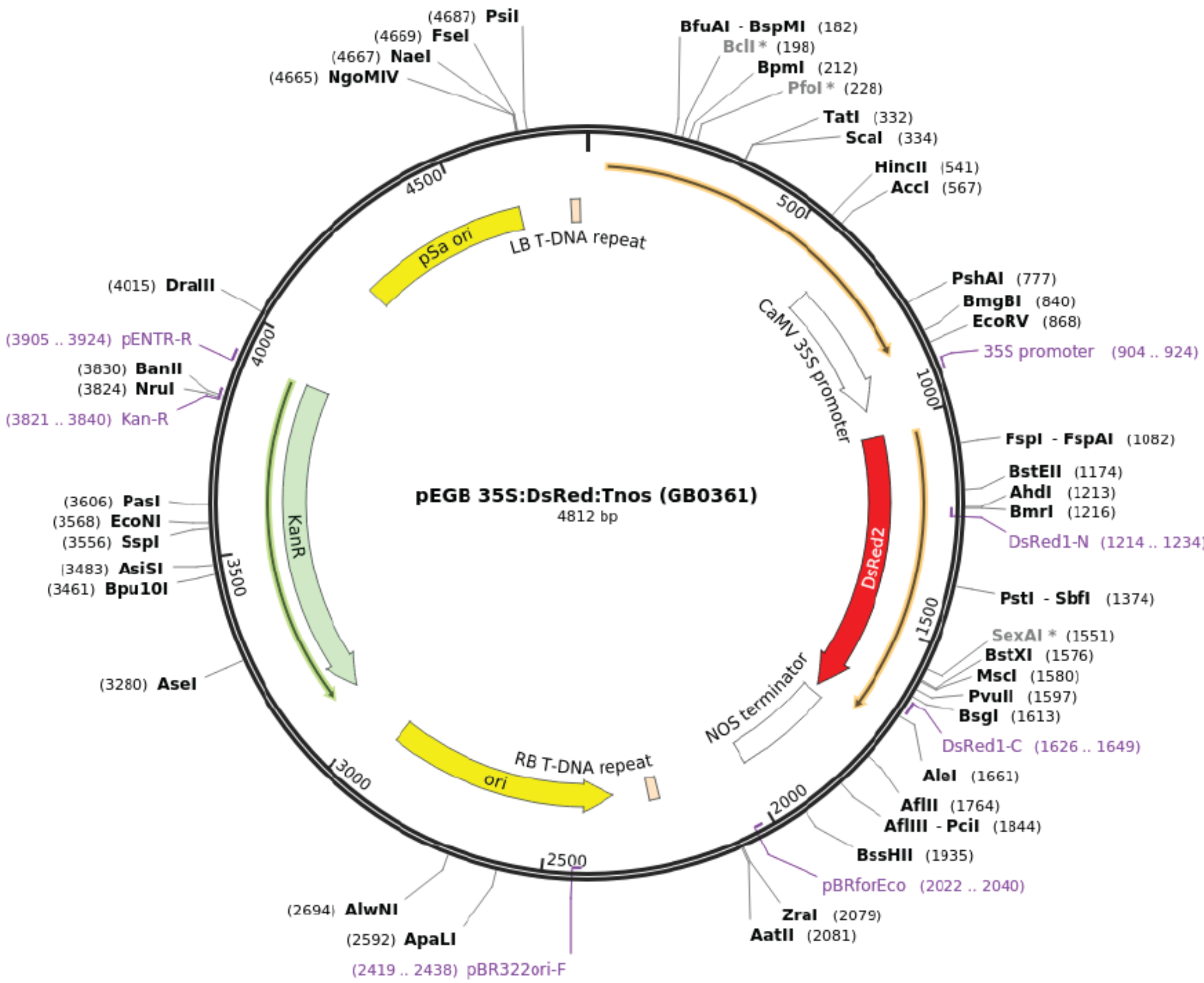
```
#Add Phidgets library
from Phidget22.Phidget import *
from Phidget22.Devices.VoltageInput import *
#Required for sleep statement
import time

#Create
ch = VoltageInput()

#Set addressing parameters to specify which channel to open (if any)
ch.setIsHubPortDevice(True)
ch.setHubPort(0)

#Open
ch.openWaitForAttachment(5000)

#Set the sensor type to match the analog sensor you are using after opening the
Phidget
ch.setSensorType(VoltageSensorType.SENSOR_TYPE_1130_PH)

#Record data points
count = 0

#define a variable
sensorValue = ch.getSensorValue()

#Use your Phidgets
while (True):
        #Update user
        print("Logging data...")

        #Write data to file in CSV format
        with open ('phidgets_temperature.csv','a') as datafile:
            datafile.write(str(sensorValue) + "\n")

        #Increment count
        count += 1

        #If 10 data points have been recorded, close file and exit program
        if(count == 10):
            print("Logging complete, exiting program")
            exit()

        time.sleep(0.5)
main()
```



Scheme S1. Schematic of the MOF-Gun.



Scheme S2. MOF-Gun nozzle



Scheme S3. Restriction map and multiple cloning sites of pEGB 35S:DsRed:Tnos (GB0361) provided by Addgene (USA) and amplified in the laboratory.

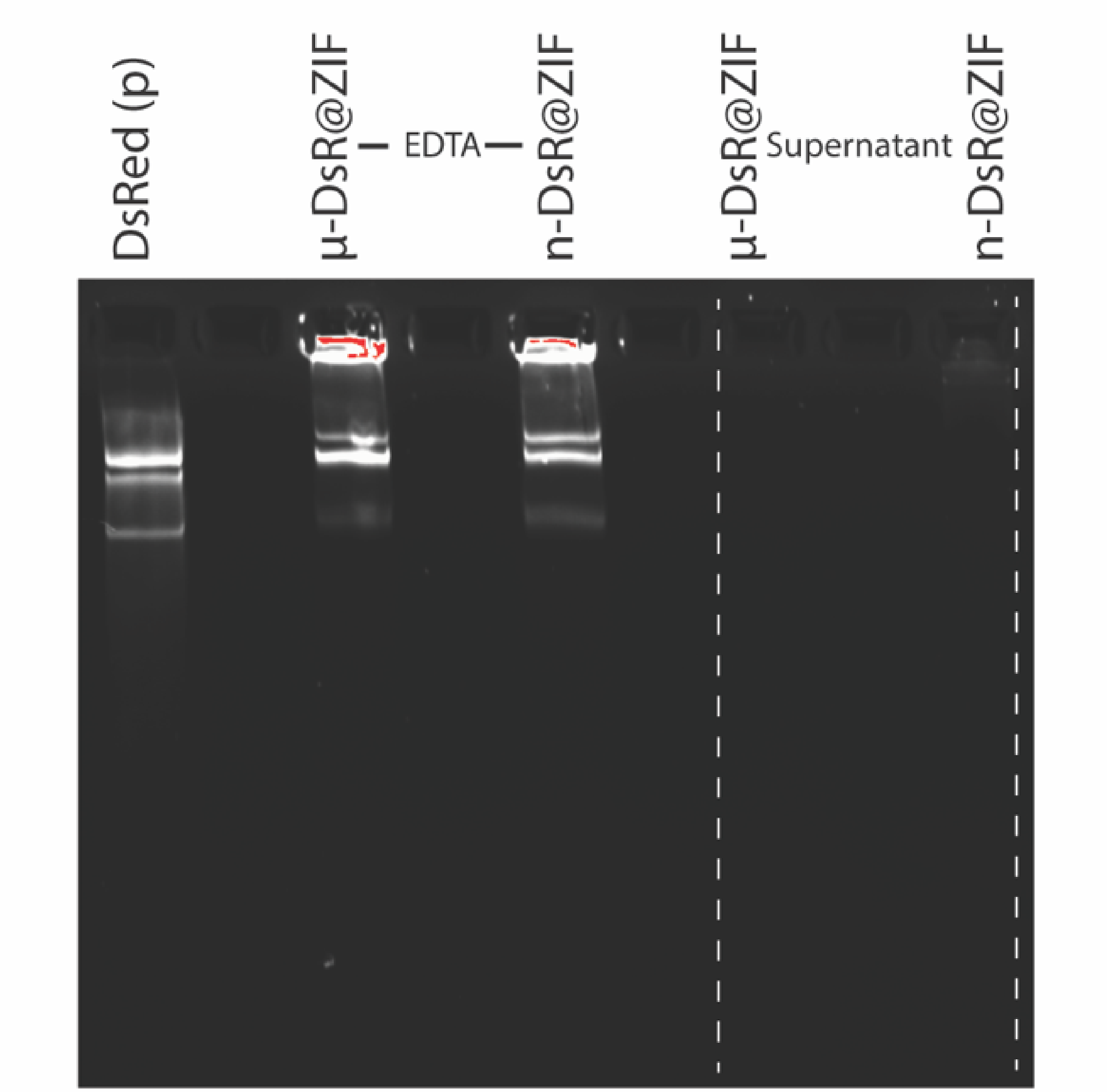


Figure S1. Agarose gel electrophoresis of DsRed (p), exfoliated micro and nano DsR@ZIF-8 (with EDTA), and the supernatants of micro and nano DsR@ZIF formulations.

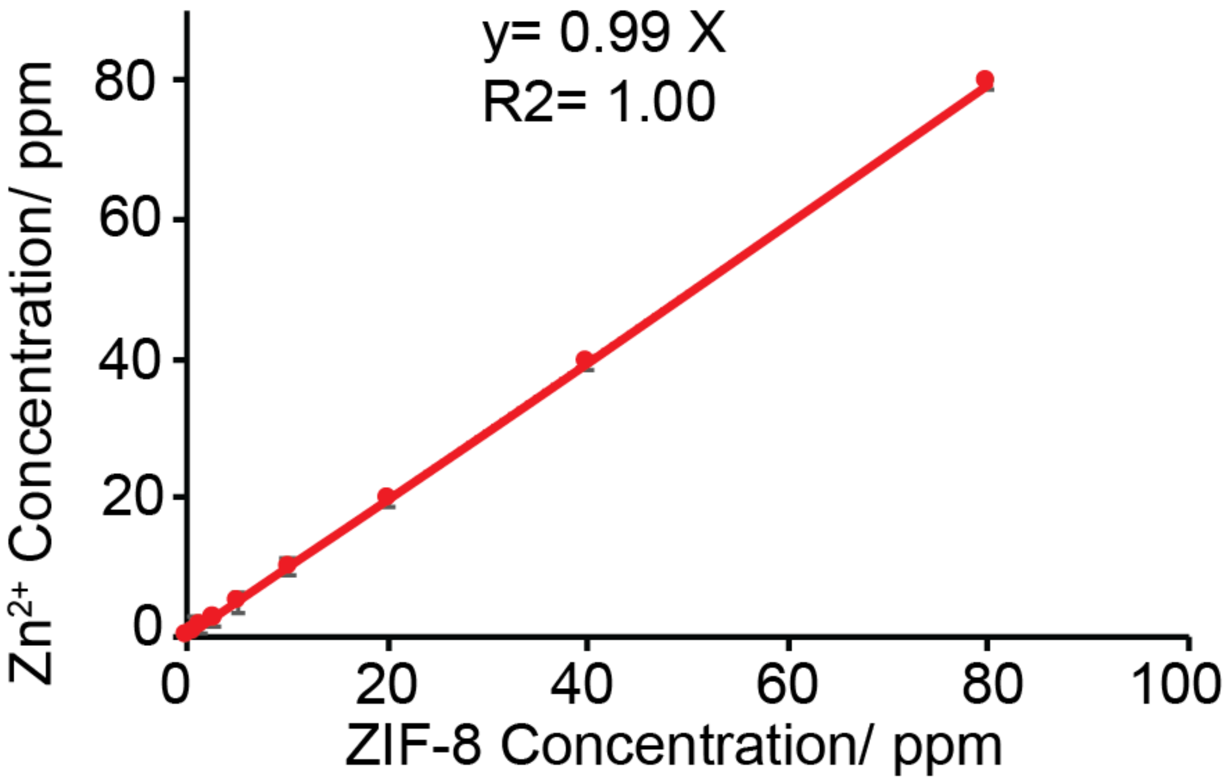


Figure S2. Standard curve for Zn^{2+} concentration used in ICP-MS.

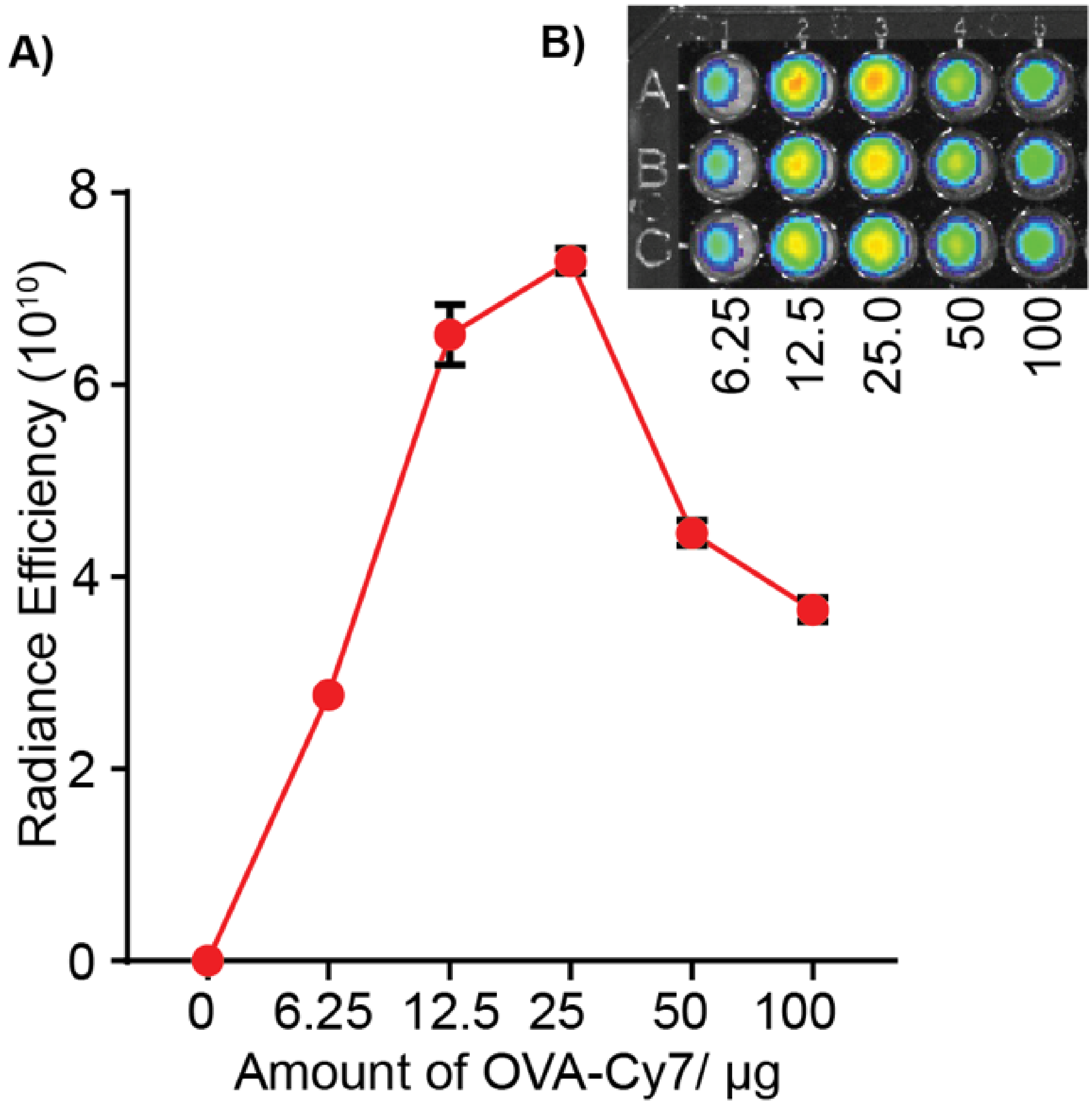


Figure S3. Radiance efficiency data was recorded from the animal imager for 96-well plate settings to determine the optimum fluorescence with different amounts of OVA-Cy7 encapsulated in ZIF-8.

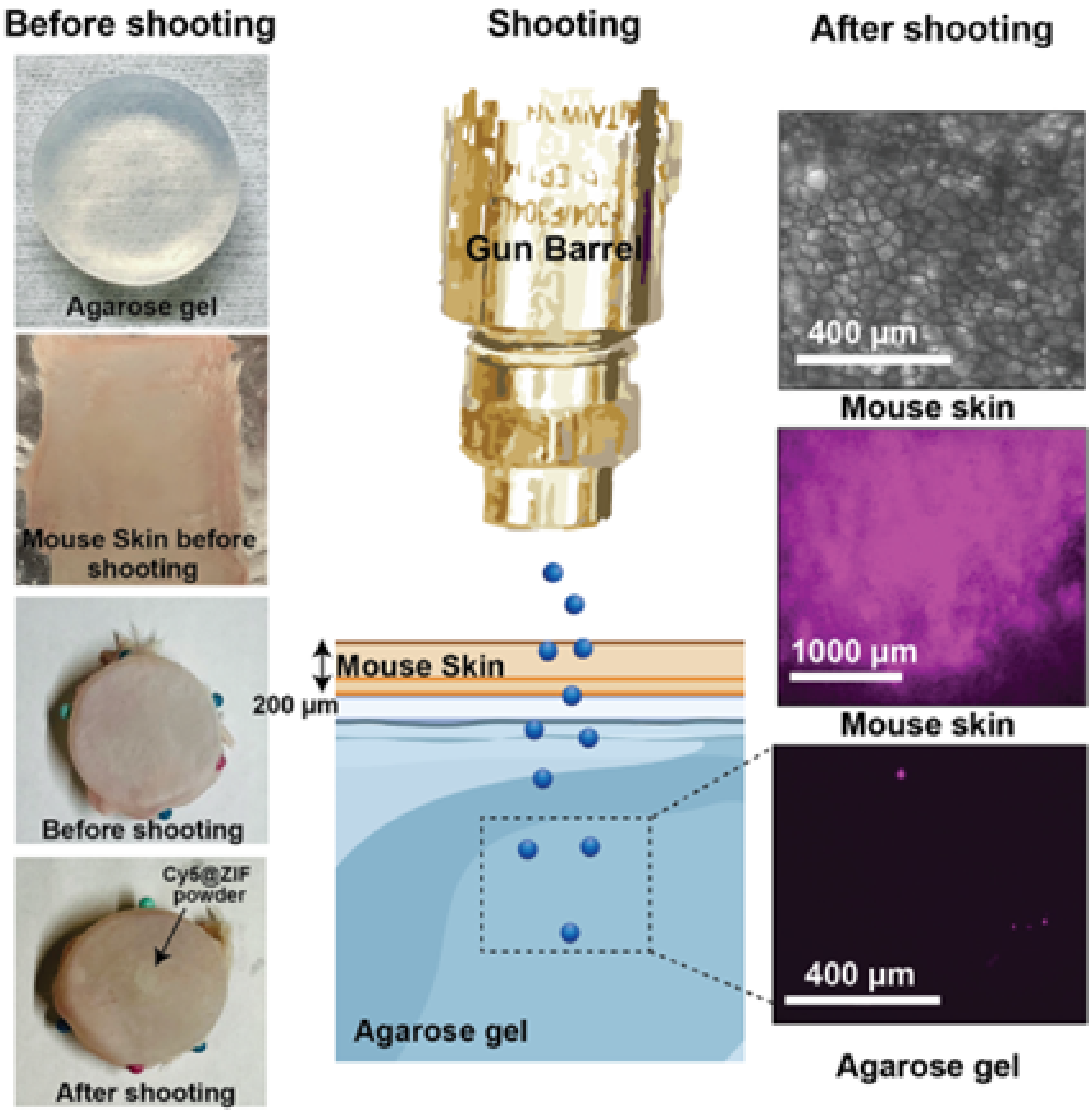


Figure S4.A-E) Optimization experiment for i*n vivo* protein release experiment. Pressure used was 500 PSI, the Cy5@ZIF penetrated the gel through the skin-covered ballistic gel. F) bright field microscopic image of the skin after shooting. G) Fluorescence image of the skin after shooting. I) fluorescence image of the gel after shooting and removing the skin from the gel surface.

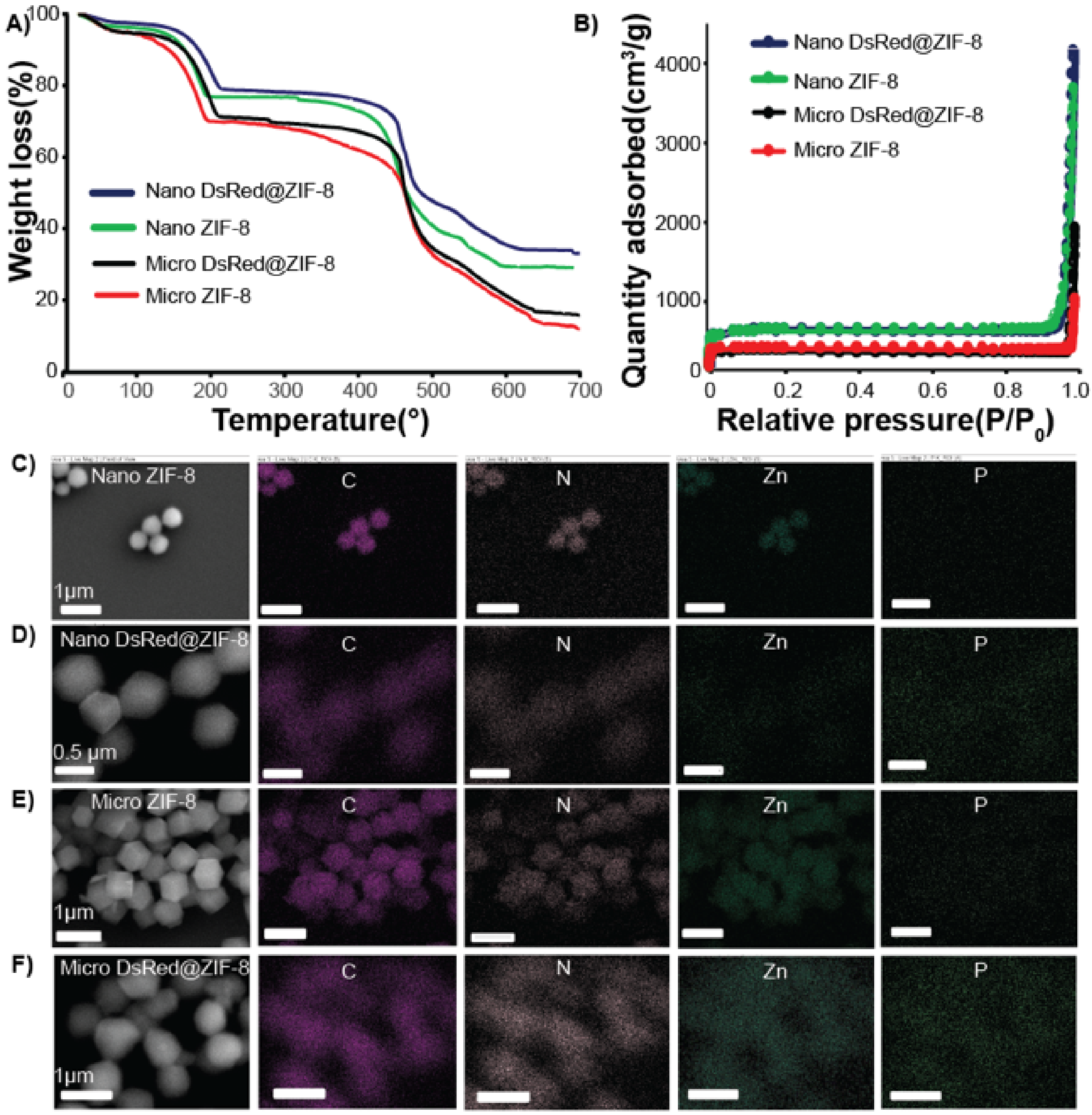


Figure S5. Characterization data was obtained from (A) thermogravimetric analysis of DsR@ZIF biocomposites along with pristine ZIF-8 controls (B) N_2 adsorption isotherm/BET data of DsR@ZIF biocomposites along with pristine ZIF-8 controls. EDAX of (C) nano ZIF-8 (D) nano DsR@ZIF € micro ZIF-8 (F) micro DsR@ZIF for elemental analysis of Carbon-C, Nitrogen-N, Zinc-Zn, and Phosphorous-P.

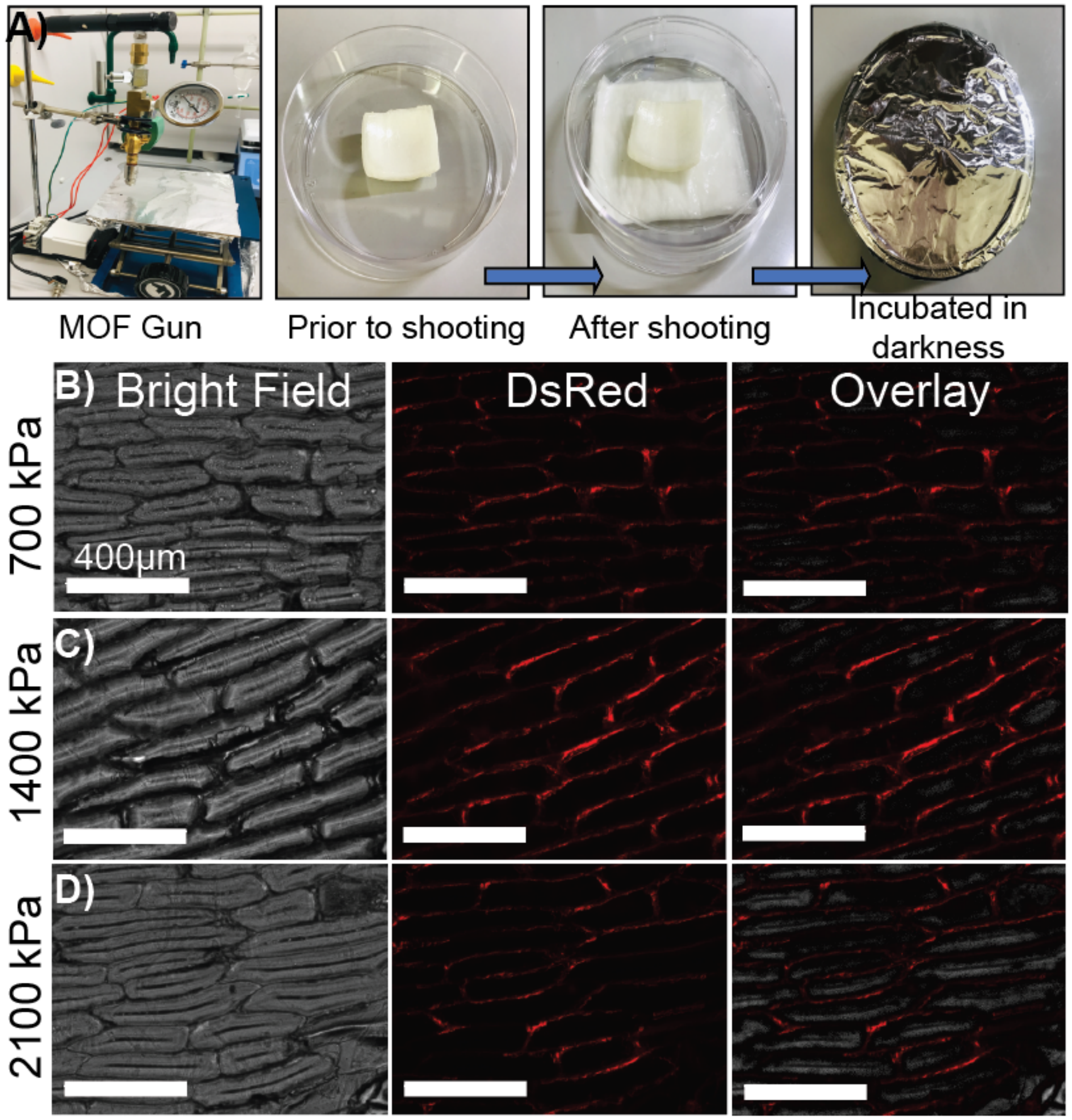


Figure S6. A) Process of gene transfection showing shooting DsR@ZIF into an onion bulb and then incubating in dark conditions for 24 h. B) Confocal images of DsRed fluorescent protein expression with DsR@ZIF with different pressure values.

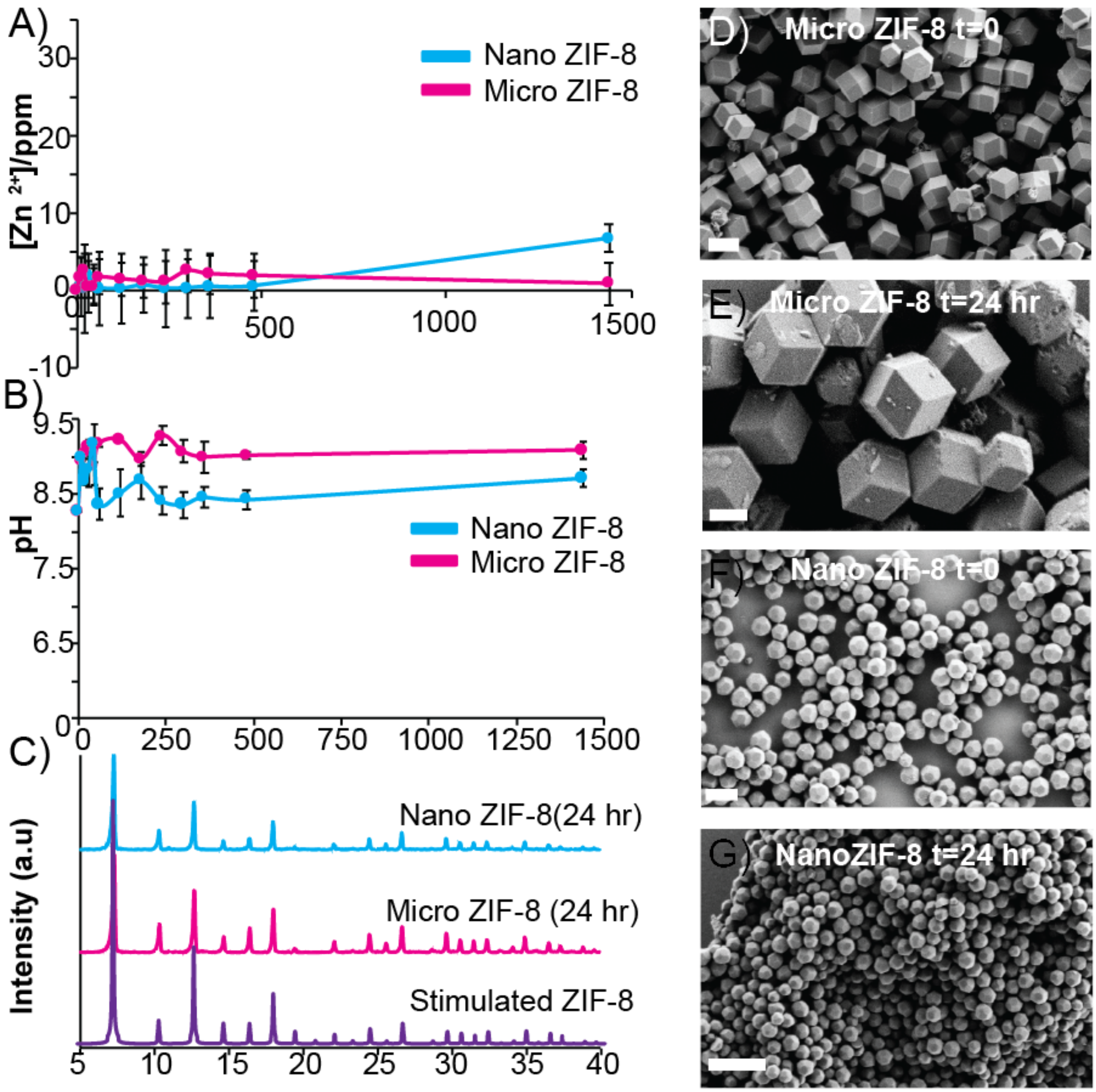


Figure S7. A) The Zn^{2+} releasing profile of ZIF-8 resuspended in DI water without shooting. B) pH profile of the solution of ZIF-8 in DI water. C) PXRDs of the micro and nano ZIF-8 were obtained after 24 h in DI water. SEM micrographs of micro ZIF-8 at D) t=0 and E) t= 24 h. Nano ZIF-8 at F) t=0 and G) t=24 h. Scale bars= 1 µm.

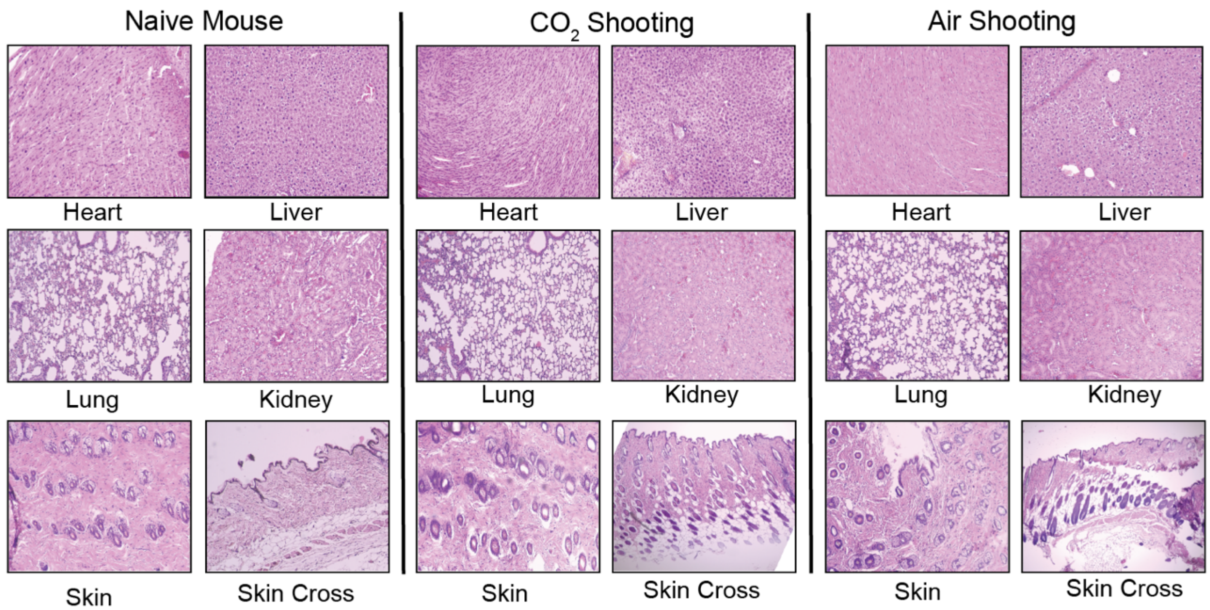


Figure S8. A) H&E staining of the major organs and skin tissues of male BALB/c mice after the shooting experiment of smURFP@ZIF-8 using 3450 kPa pressure, 1 cm shooting distance from the gun nozzle to the skin surface.

Table S1. Ingredients used for the synthesis of micro ZIF-8 formulations.

Formulation	3 M mIM volume (mL)	1 M zinc acetate volume (mL)	Nuclease-free water volume (mL)	DsRed(p) volume (mL)
Micro ZIF-8	0.213	0.010	0.777	0
Micro DsR@ZIF	0.213	0.010	0.752	0.025

Table S2. Ingredients used for the synthesis of nano ZIF-8 formulations.

Formulation	mIM weight (mg)	Zinc acetate weight (mg)	Nuclease free water volume (mL)	DsRed (p)/ OVA-Cy7 volume (mL)
Nano ZIF-8	95.0	7.0	1.000	0
Nano DsR@ZIF	95.0	7.0	1.000	0.025
Nano OVA-Cy7@ZIF	95.0	7.0	1.000	0.003

Table S3. Optimization of biolistic parameters for plant gene delivery

Distance from the nozzle/ cm	Penetration depth under different pressures/ mm		
	700 kPa	1400 kPa	2100 kPa
1	0.5	3.0	X
2	0.4	2.5	1.5
3	0.35	1.0	1.4
4	0.60	1.5	1.7