Supplementary

1. Bench model description

A bench model was developed to simulate needle delivery and measure GS deflection. The plexiglass sheet had a laser-cut grid of 1/8" holes at 1 cm intervals, 3D printed clips fitted to the holes to guide the GS shaft on the grid, and 3D printed 2 cm diameter spherical targets. Images of the GS were captured using a digital single-lens reflex camera perpendicular to the platform, and GS deflection and angles and errors were digitally measured with ImageJ (NIH, Bethesda, MD, USA).

2. *Ex vivo* lung model description

GS navigation and needle deliveries were conducted in an inflatable plasticized swine lung (BioQuest® Inflatable Lung Kit, Nasco, Fort Atkinson, WI). Gel water beads, a cross-linked super absorbent crystal polymer (Magic Beadz, Sanavet Laboratories LLC, US), absorb liquid and swell in size to an average solid sphere size of 6 ± 2 mm after injection in the lung (n=5). Beads were injected in the lung transpleurally through a 6 Fr introducer sheath to serve as surrogate lung targets. The puncture sites were sealed with a drop of liquid bandage. The lung was placed in a sealed container and mechanically ventilated using a vacuum pump connected to the container. GS navigation and needle delivery to each target were performed with CBCT-based AF image guidance.

3. Swine management and anesthesia details

Anesthesia: Animals were sedated with intramuscular ketamine (25 mg/kg), midazolam (0.5 mg/kg), and glycopyrrolate (0.01 mg/kg) and anesthetized with Propofol (1 mg/kg IV). Animals were intubated, maintained under general anesthesia with isoflurane, 1–5% (Isoflo, Abbott Animal Health; North Chicago, IL), and mechanically ventilated with breathing temporarily suspended during CT or CBCT acquisition. At the conclusion of the study, euthanasia was performed under general anesthesia by intravenous administration of Beuthanasia-D (pentobarbital sodium 390 mg/mL and phenytoin sodium 50 mg/mL).
Swine Housing conditions: The Division of Veterinary Resources (DVR) facility set point for swine is 72º F +/– 3º F. The DVR facility set point for humidity is 50%. Animals have outdoor access when temperatures are >32*F and <90*F.
Animal Enrichment: Pigs are socially housed and provided with toys to encourage normal behaviors. Animals are provided with fresh vegetables and foraging materials. All animals were enriched and housed in groups prior to experimentation.
Animal Health Status: Animals come from a closed colony and the herd is free of PRRS, PRV, Brucellosis, Myocoplasma hypopneumoniae, and APP. Animals are vaccinated with an autogenous vaccine consisting of Streptococcus Suis, Mycoplasma Hyorhinis, Mycoplasma Hyosynoviae, and Haemophilus Parasuis, in addition to commercial vaccinations for swine influenza, Bordetella bronciseptica, porcine circovirus, Erysipelas rhusiophathiae and Salmonella Choleraesuis-Typhimurium.
Diet: Ziegler NIH Swine 2004-2.

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