[Honeybee] In vivo gene transfer into the adult honeybee brain by using electroporation



micropipette

С

In vivo electroporation.

- (A) The necks of honeybees that had settled in the tube were fixed in place by inserting two U-shaped plastic sheets for surgical ease and later release.
- (B) A honeybee settled in the tube with her neck fixed prior to the operation.
- (C) Schematic representation of the micropipette and electrodes placed in the honeybee brain. Purple indicates micropipette filled with DNA solution, blue indicates electrodes. AN, antennae; C, compound eyes; MB, mushroom bodies; Oc, ocelli; OL, optic lobes.
- (D) DNA injection and electroporation with needle electrodes.

Arrows and arrowhead indicate electrodes and micropipette for DNA injection, respectively. Scale bar, 300 m.



Fluorescent images of electroporated honeybee brain at 50 V, targeting the optic lobe (A, B) and mushroom bodies (C-F).

Fluorescent signals were detected around the anode position in the brain electroporated with GFP-expressing plasmid driven by the CMV promoter (A, C, E, F) and not in those electroporated without plasmid (B, D).

(G, H) Magnified views of GFP-expressing areas shown in (A) and (F), respectively.

Positions of electrodes are indicated by circles containing + or -.

Arrowheads indicate the regions in which fluorescence was detected (A, C, E, G) and neural projections from mushroom body cells (F, H).

Frontal view (A, B, F, G, H), rear view (C, D), and top view (E). Panels (C) and (E) show different views of the same brain.

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