CATALOG

Supplemental Results

Preliminary results establishing a new blood-brain barrier permeability evaluation method

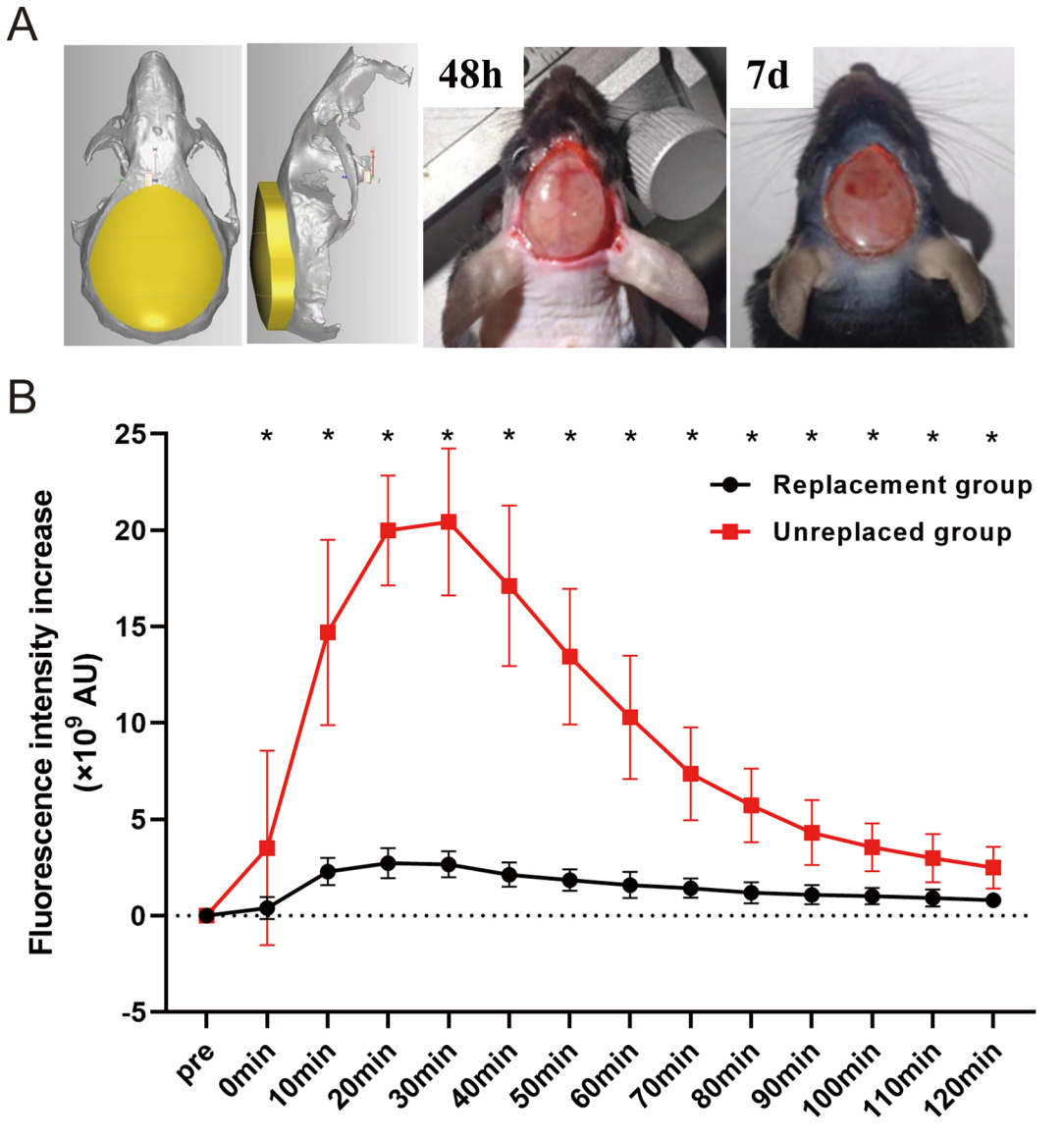
For generalized and progressive blood-brain barrier lesions, we set the examination area to cover the entire brain as much as possible. After a single injection of sodium fluorescein, we observed a significant difference in the trend of fluorescence signal changes in the region of interest of mice that underwent skull transparency through this method compared to mice that did not undergo skull flap replacement surgery ($F = 144.889$, $P = 0.000$). Figure S1B illustrates that the intragroup data were more consistent and avoided the continuous signal interference caused by the low vascular permeability of skin and connective tissue, which could result in fluorescence molecules entering the tissue. This highlights the importance of achieving skull transparency before conducting body imaging.

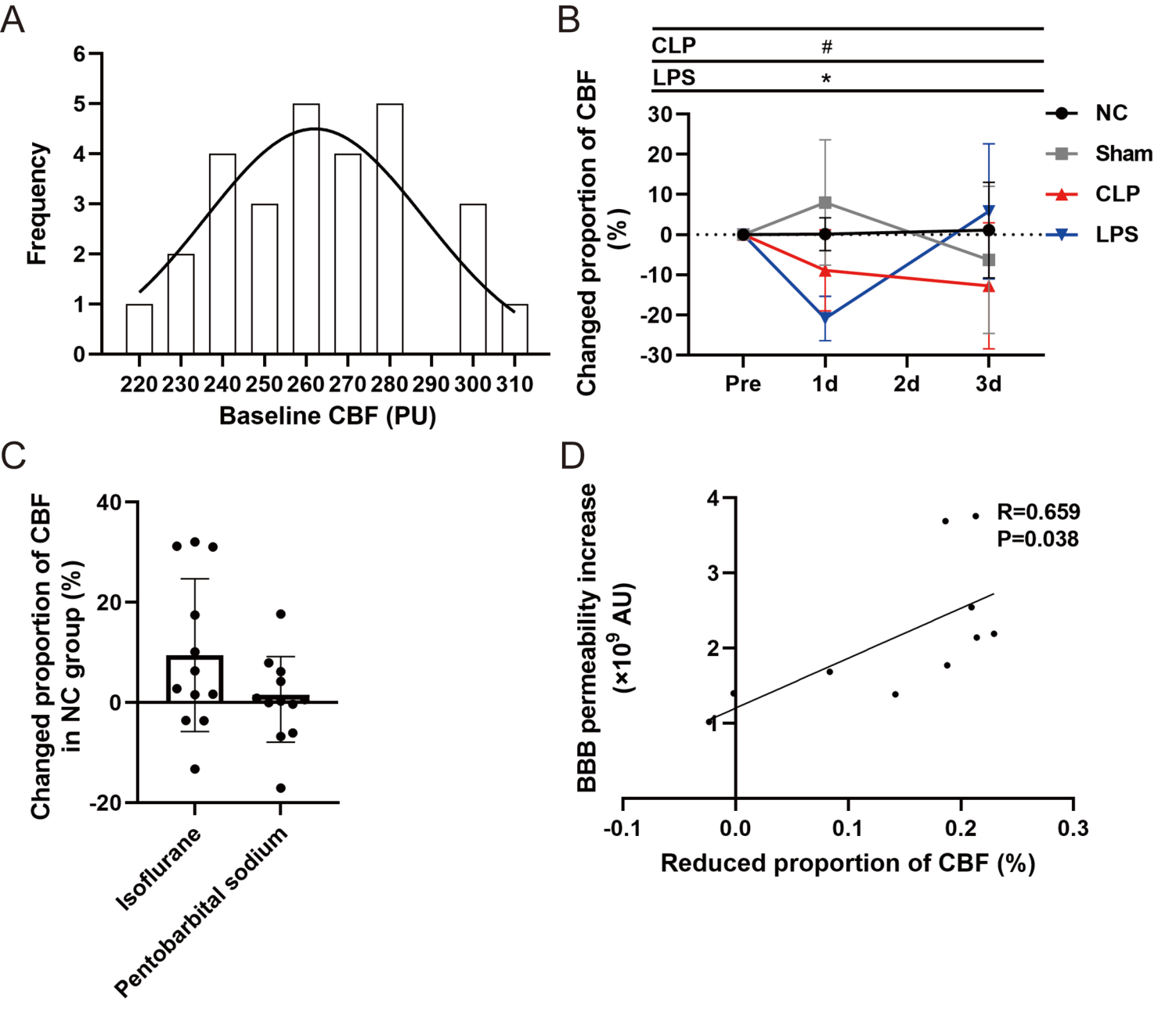
Considering the standardization and skull integrity requirements, we designed a cap-shaped cover plate and a flat cover plate based on the scanned mouse skull shape, and discarded ground cover slides as shown in Figures 2B and S1A. During skull cover plate replacement and in vivo fluorescence imaging, it was found that the cap-shaped cover plate had defects including fluorescence reflection interference during imaging caused by polishing the curved surface and residual air bubbles that often occur during surgery because of the close proximity of the structure to the skull. Furthermore, too little adhesive under the cover plate can lead to severe connective tissue hyperplasia 7 days after surgery (Figure S1A). Applying a flat cover plate can prevent the above problems and ensure the feasibility of the new method.

Supplemental Figure Legends

Figure S1. Verification of the feasibility and necessity of skull cover plate replacement for in vivo fluorescence imaging: (A) Typical images of replacement position and maintenance effect of a flat cover plate; (B) Exploring the effect of skull cover plate replacement on the fluorescence dynamics of brain tissue in regions of interest at the same location in mice, after a single injection of sodium fluorescein. '*' vs the replacement group, $P < 0.05$.

Figure S2. The changing trend of cerebral microcirculation perfusion volume and the effect of anesthetics on cerebral blood flow in mice after establishment of a sepsis model: (A) The baseline cerebral blood flow distribution of mice; (B) Use of pentobarbital sodium anesthesia to monitor the cerebral blood flow change trend with time in each mice group within 3 days after modeling; (C) Under pentobarbital sodium and isoflurane anesthesia, the proportion of changes in cerebral blood flow of healthy mice within 24 h before and after anesthesia were compared; (D) The fitting curve obtained by linear correlation analysis between the proportion of cerebral blood flow decrease at 12 h and the quantitative value of blood-brain barrier permeability increase at 24 h after modeling, measured under isoflurane anesthesia. '*' $P < 0.05$; '**' $P < 0.01$.


A
48h
7d
B
Fluorescence intensity increase
(×10^9 AU)
Replacement group
Unreplaced group
25
20
15
10
5
0
-5
*
pre
0min
10min
20min
30min
40min
50min
60min
70min
80min
90min
100min
110min
120min


A
Frequency
Baseline CBF (PU)
B
CLP
LPS
#
*
Changed proportion of CBF (%)
NC
Sham
CLP
LPS
Pre
1d
2d
3d
C
Changed proportion of CBF in NC group (%)
Isoflurane
Pentobarbital sodium
D
BBB permeability increase (×10^9 AU)
R=0.659
P=0.038
Reduced proportion of CBF (%)