Cysteine Nitrosylation of Adenylyl Cyclase Isoform 6 and Forskolin Rescue in Hypoxic Pulmonary Hypertension of the Newborn

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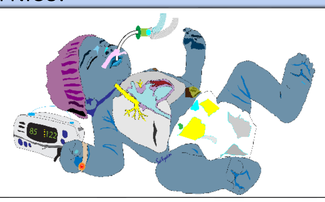
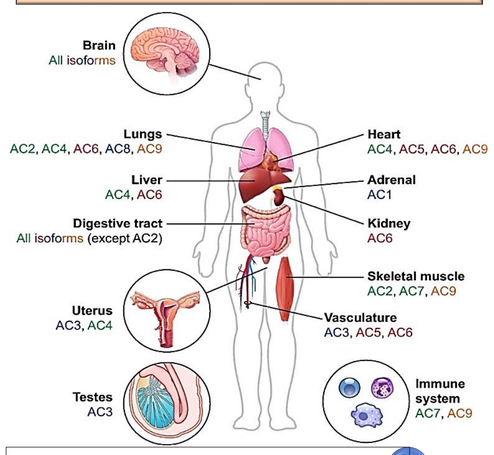
Background

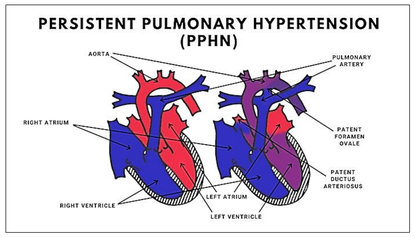
Persistent Pulmonary Hypertension of the Newborn (PPHN) is a catastrophic respiratory failure of the normal pulmonary vascular relaxation after birth. These blue babies are the sickest in NICU.

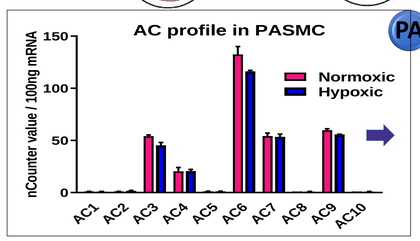


PPHN is characterized by Pulmonary vasoconstriction causing Hypoxemia secondary to right-to-left shunting of blood at the foramen ovale and ductus arteriosus.

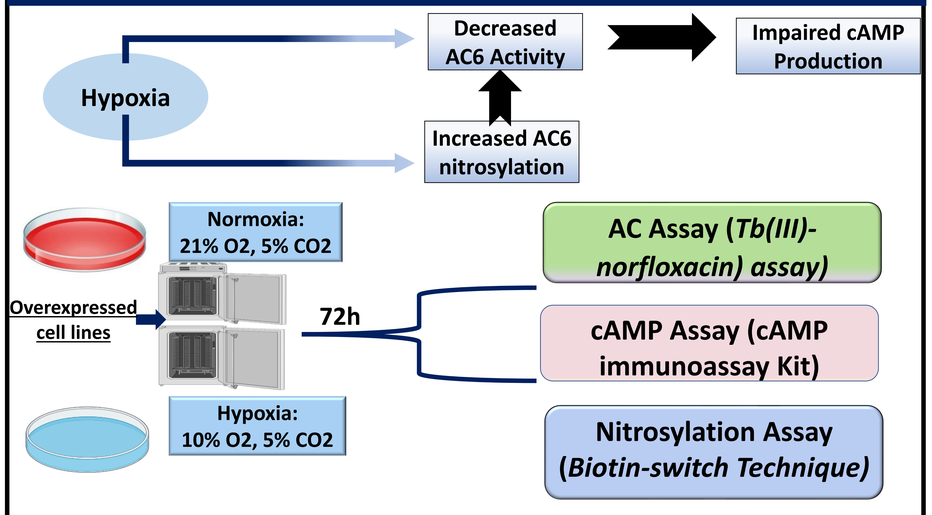
Our previous results show that in pulmonary arteries, **AC6** is predominant, followed by AC3, AC7 and AC9







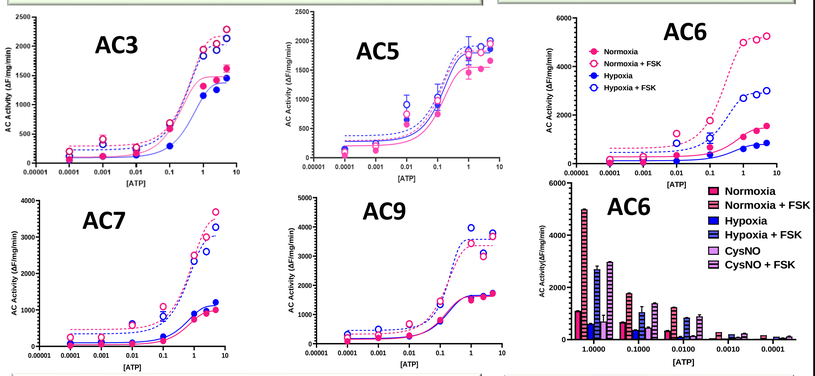
Methods



Results

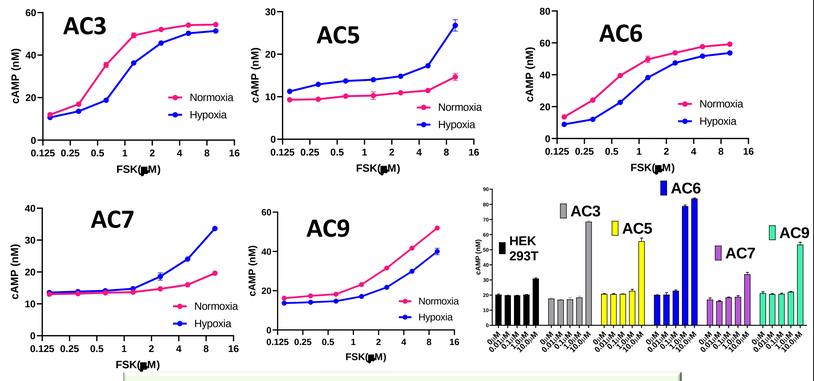
Among AC isoforms tested, only AC6 is inhibited by hypoxia and only AC6 is inhibited by nitrosocysteine (CysNo) (NO donor).

AC3, 5, 6, 7 & 9 are activated by FSK: activation of AC6 is impaired in hypoxia.



Hypoxia impaired cAMP production by AC6, and to some extent by AC3 and 5. For all AC isoforms, FSK increased cAMP generation.

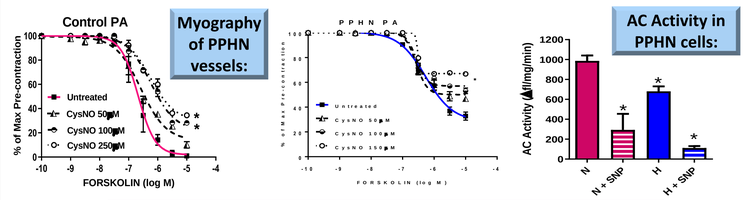
Compared to all AC isoforms, AC6 was highly sensitive to FSK



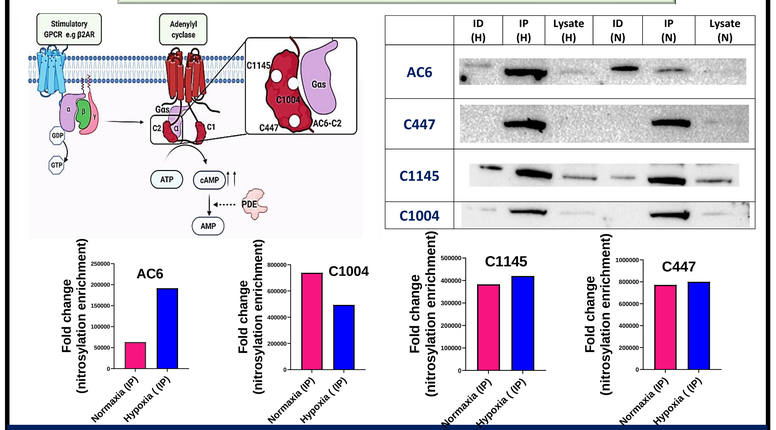
Hypoxia Significantly Increased AC6 Nitrosylation

Results

Previous Data from our lab: CysNO and sodium nitroprusside (SNP) inhibited FSK-mediated relaxation of PPHN and control pulmonary arteries. Hypoxia and SNP decreased AC activity in PPHN cells



Which Cysteine Residues of AC6 are Nitrosylated?



Conclusions

- **Hypoxia leads to decreased AC6 activity – but not AC3, AC5, AC7 or AC9**
- **Hypoxia leads to decreased AC6 cAMP production, but it can be rescued with FSK**
- **Hypoxia increases nitrosylation of AC6, but not that of AC3, AC5, AC7 or AC9**
- **Nitrosylation decreases AC6 activity, but not that of other AC isoforms.**
- **Cys1004 in AC6 is present at the AC–Gαs interface. Substitution mutation of Cys1004 suggests that this is the residue nitrosylated under hypoxic conditions.**

Future Directions

- **AC6 is a therapeutic target in PPHN. If cysteine nitrosylation is the mechanism responsible for inhibition of AC6 activity by hypoxia, we could design derivatives of the FSK parent compound that are selective for AC6, to reactivate a hypoxic AC6. This may inform the development of new preventative and therapeutic strategies against PPHN.**