고압산소치료와 기초 및 임상연구

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내용

- 현재: 동물연구
  세포연구
  임상연구
  챔버연구
- 미래: 향후연구

내용

- 연구의 결과는 원주의대 고압의학연구소 팀원의 결과물입니다

고압산소치료란?
(HBO_2: hyperbaric oxygen therapy)

- 정의: 챔버를 이용하여 1.4기압 이상으로 100%산소를 호흡시킴으로 의학적 효과를 나타내는 치료법 (의학적 압력은 2.0 – 3.0 기압)

- 원리와 효과 – 고농도 산소 유지(hyperoxygenation)

Approved Ixs for HBO_2 therapy (14th E)

Disorder	HBO_2 mechanism	Typical dose	Utilization review	Level of evidence	Class of recommendation
Air or gas embolism	Bubble reduction	U.S. Navy Table 6 or equivalent	10	C-LD	I
CO	CO washout	Up to 3 ATA 1-3 sessions or to clinical plateau	3	A	IIa
DM foot	Treat ischemia	2-2.5 ATA Daily for 3-4 weeks, based on healing response	30	A	I
SSNHL	Supraphysiologic levels of O_2 in tissue	2-2.5 ATA 10-20 sessions	20	A	IIa
Thermal burn	Inflammation modulation	2-2.4 ATA Twice daily Up to 30 sessions	30	A	IIa

연구주제

- 압력: optimal pressure, frequency, duration
  (각 적응증에 따른 동물, 임상 연구)
- 새로운 적응증
- 외국인 vs 한국인

연구하드웨어

- 한국형 고압산소치료 프로토콜 개발
- 임상연구

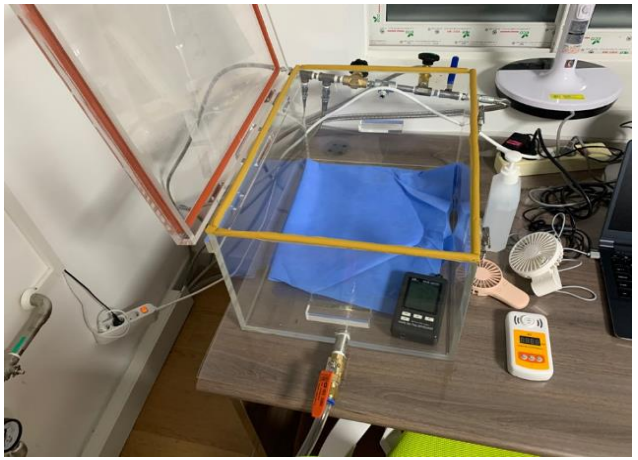
- 치료 메커니즘
- 미토콘드리아 효과

- 동물/세포실험용 고압산소 치료기
- 동물/세포 연구

동물실험연구

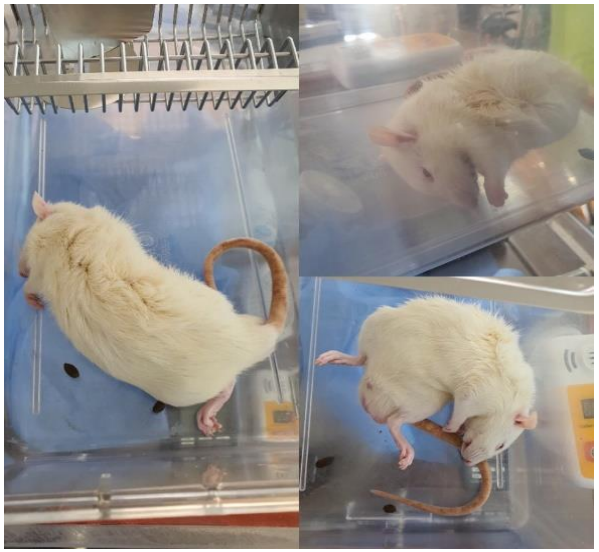
0 동물용 고압산소챔버

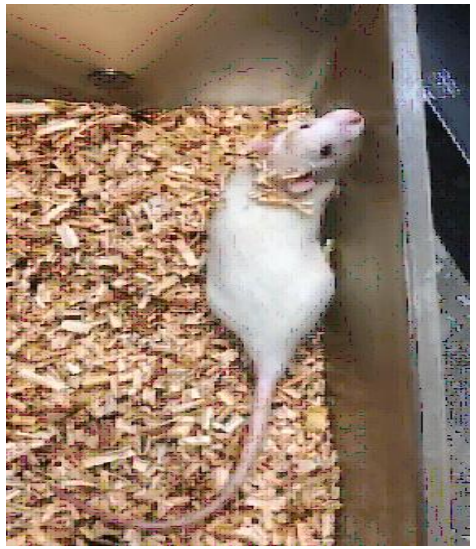
- 동물실험용 고압산소챔버 설치 완료
- 동물실험용 고압산소챔버 가동을 위한 공간 확보 및 기기설치 완료
- 동물실험용 고압산소챔버 가동 및 실험 진행
- 일산화탄소 중독 실험을 위한 시설 구비 완료



동물실험연구

- 해외 선행연구 기반과 해외연구자 자문을 통하여 일산화탄소 농도 **100%** 주입 후, **1000ppm**으로 유지하여 신경손상을 유발하기 위한 실험 진행
- 첫 번째 예비실험에서 **100%** 농도 주입 후 **5**분 만에 예상과 달리 흰쥐가 사망함
- 또한, **1000ppm/3000ppm**을 동물챔버 내에 주입하였으나 실제 내부 농도가**400ppm/750ppm**으로 측정되었으며, 이에 대한 언급이 선행연구에 없었음
- 여러 차례의 예비 실험을 통해 적정한 흰쥐 모델 구축 **(**내부 **3000 ppm, 15**분 노출**)**

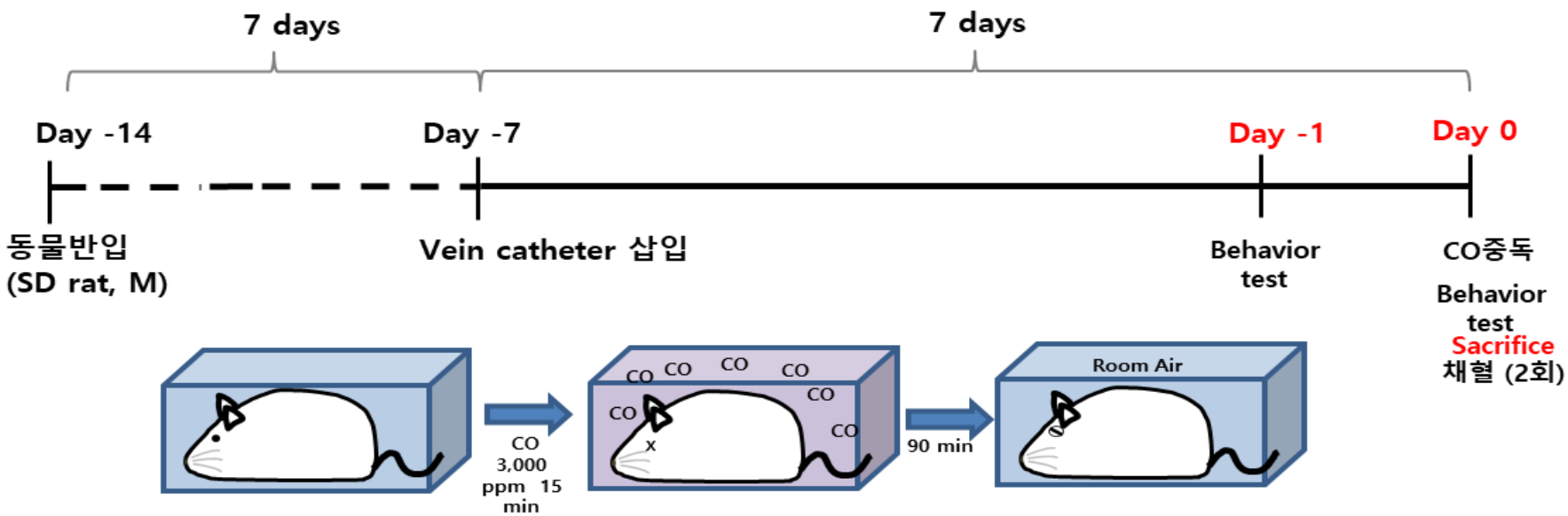




동물실험연구

1 흰쥐 급성일산화탄소중독 모델 제작

- 중독 1주일 전 혈중 gas농도를 측정하기 위한 catheter 삽입
- 중독 전 behavior test (Plus-Maze, Open field test)
- Chamber 내부 CO농도 3000ppm 으로 15분 간 노출 후 LOC확인
- 중독 90min 후 behavior test
- Sacrifice - Brain 손상 범위 확인



동물실험연구

1 흰쥐 일산화탄소 중독에 의한 LOC 유발

동물실험연구

1 흰쥐 일산화탄소 중독에 따른 혈중 CO-Hb

- 일산화탄소 중독에 의한 **LOC** 발생 시 혈중 **COHb** 농도의 정량화
- 내부 **3000 ppm, 15**분간 노출 시킨 후 **LOC** 상태에서 측정한 **CO-Hb** 값 확인
- 대기 중에서 **90min** 간 **CO wash COHb**의 변화

	Body weight	COHb(%)	O_2Hb(%)
3000ppm (15min)	350±2.06	58.53±0.82	31.03±5.49
After 90min		10.38±0.62	63.53±1.74

(Mean±SEM)

동물실험연구

1 흰쥐 일산화탄소 중독에 따른 정서-행동의 변화

- **Open Field Test**

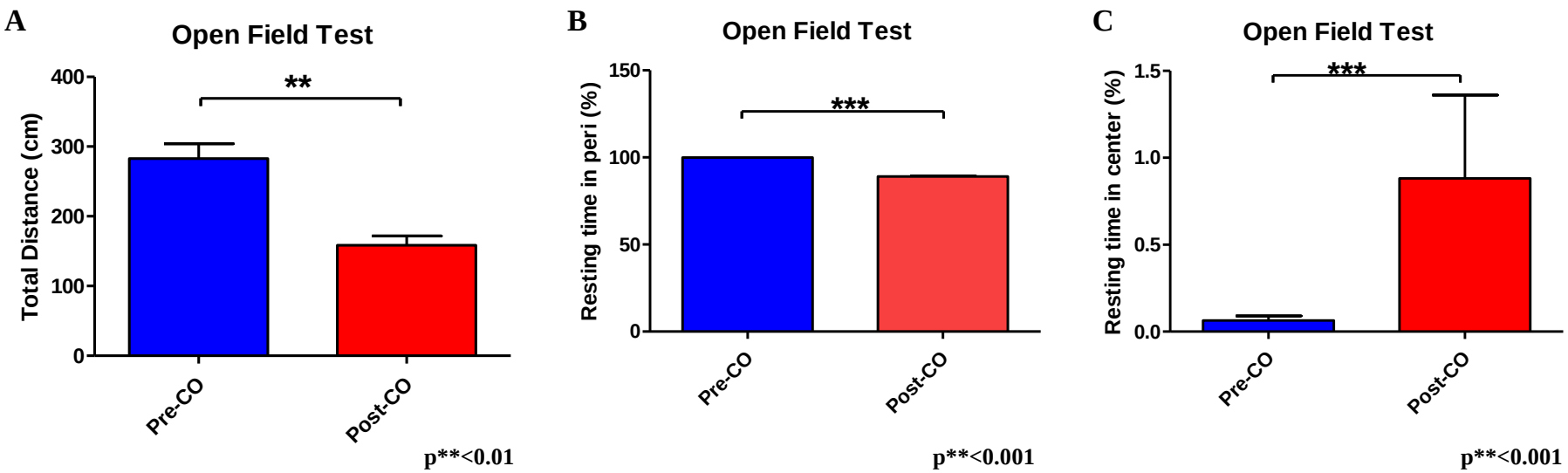
CO 중독 전

CO 중독 후

동물실험연구

1 흰쥐 일산화탄소 중독 에 따른 정서-행동 변화

- **Locomotor Activity** 감소

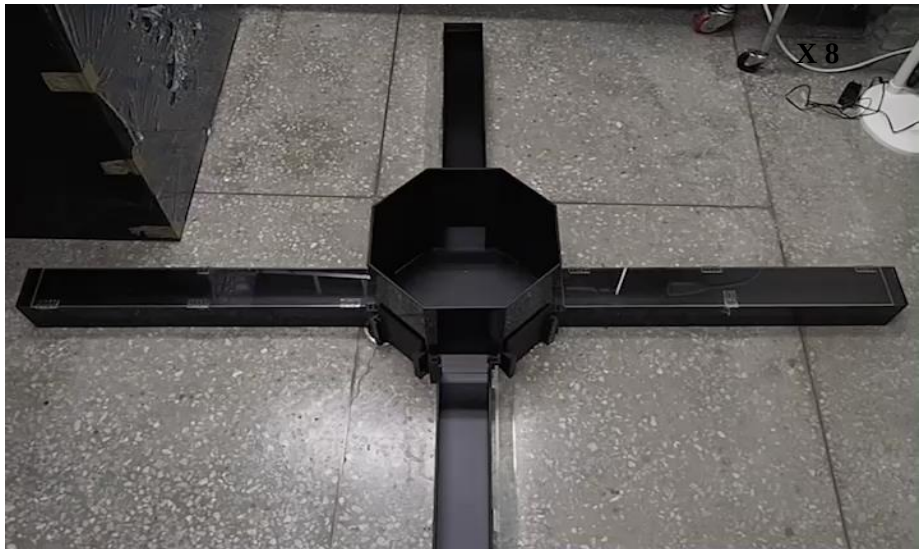


동물실험연구

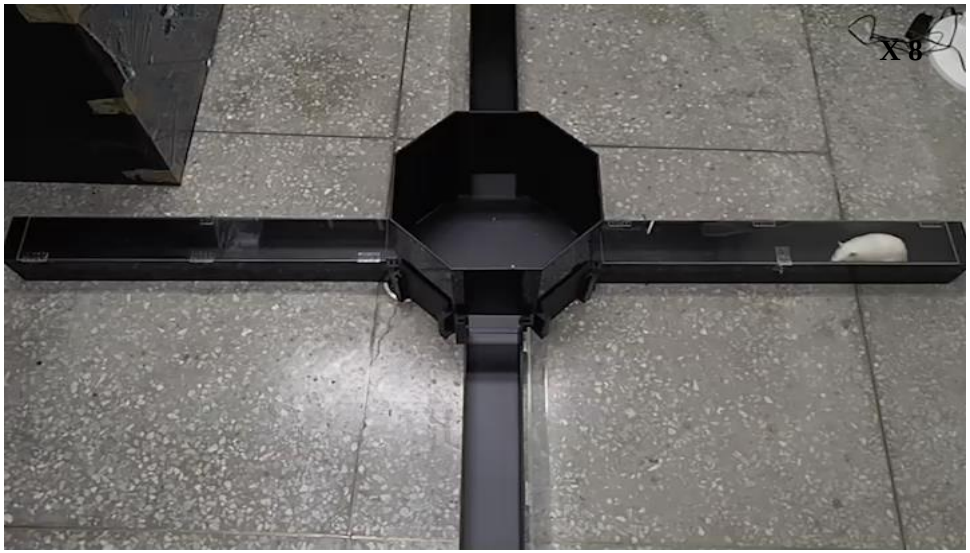
1 흰쥐 일산화탄소 중독 에 따른 정서-행동 변화

- **Plus Maze Test**

CO 중독 전



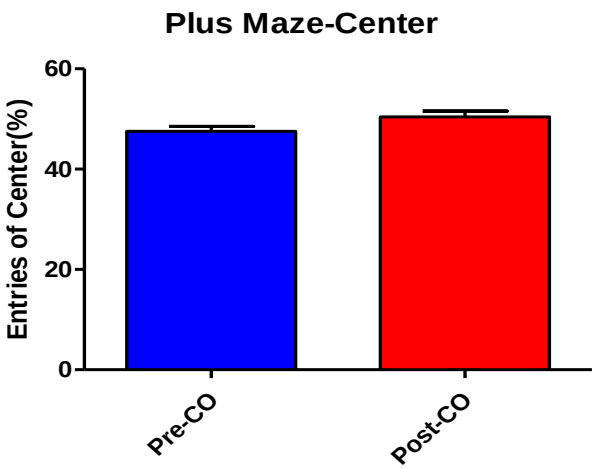
CO 중독 후

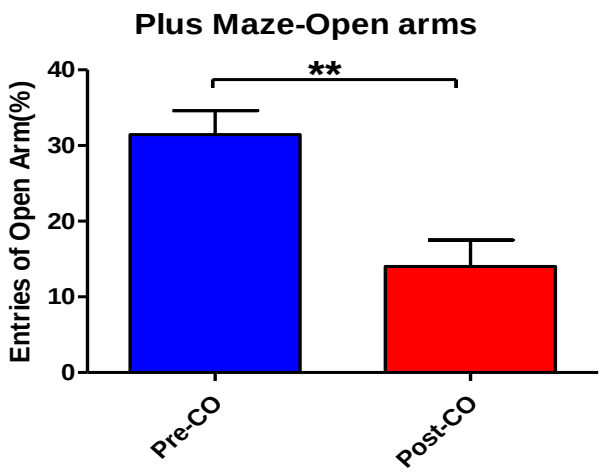


동물실험연구

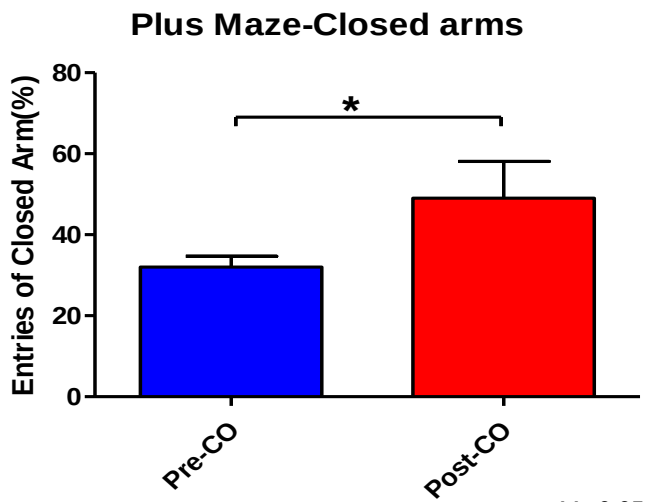
1 흰쥐 일산화탄소 중독 에 따른 정서-행동 변화

- **Behavior - Mood change (closed arm: 심리적 불안)**





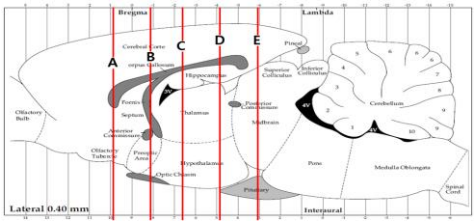
p**<0.01



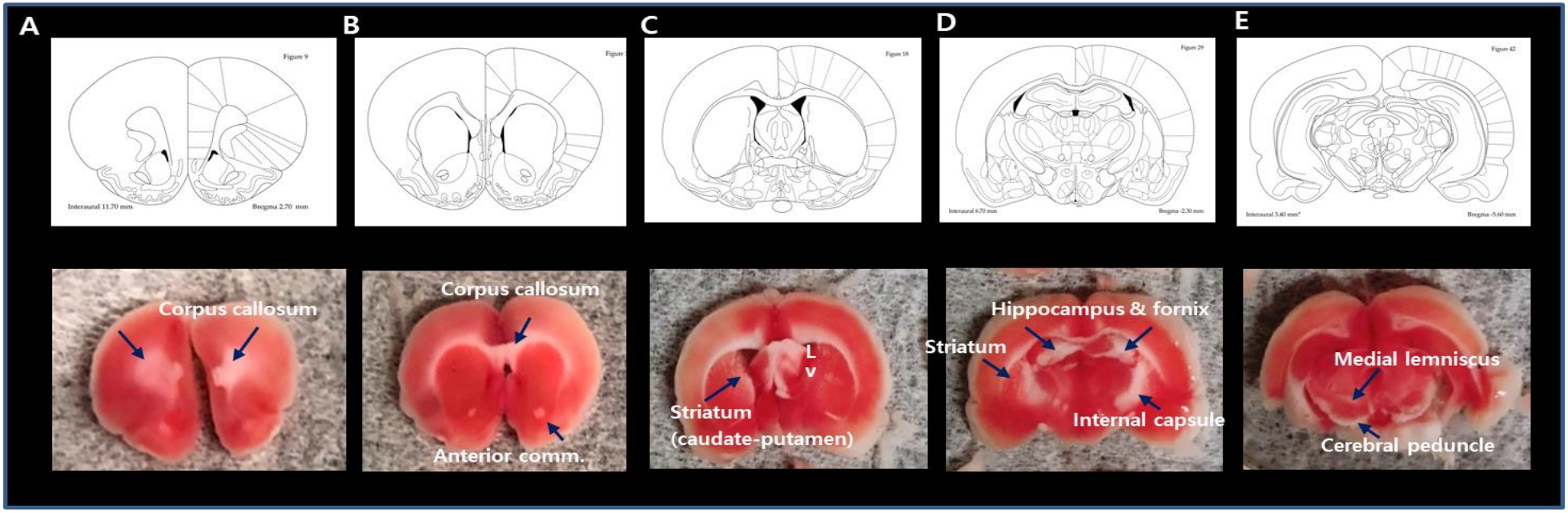
p**<0.05

동물실험연구

1 흰쥐 일산화탄소 중독 에 따른 뇌손상 검증 (명확한 손상 확인: 다른 면역염색법 진행중)



Sagittal view of rat brain

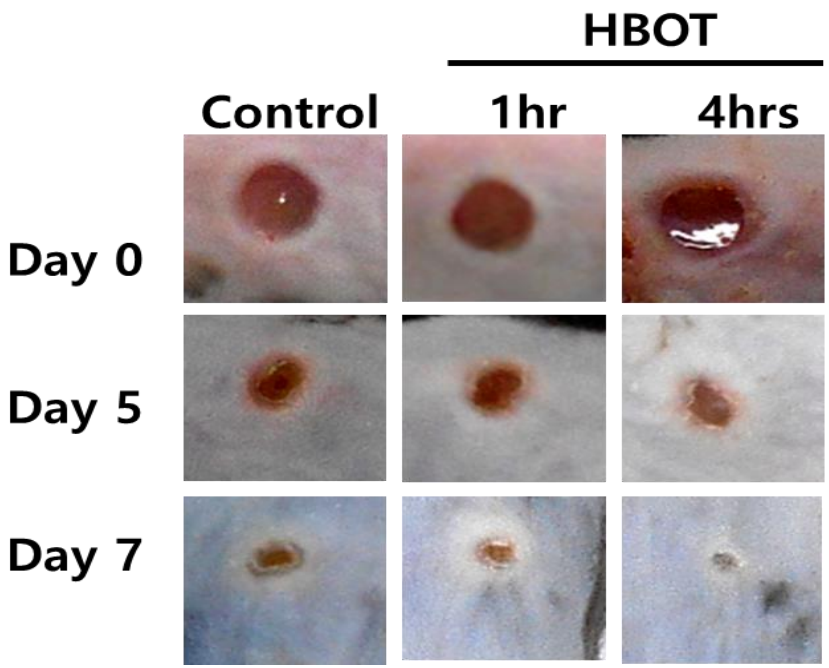


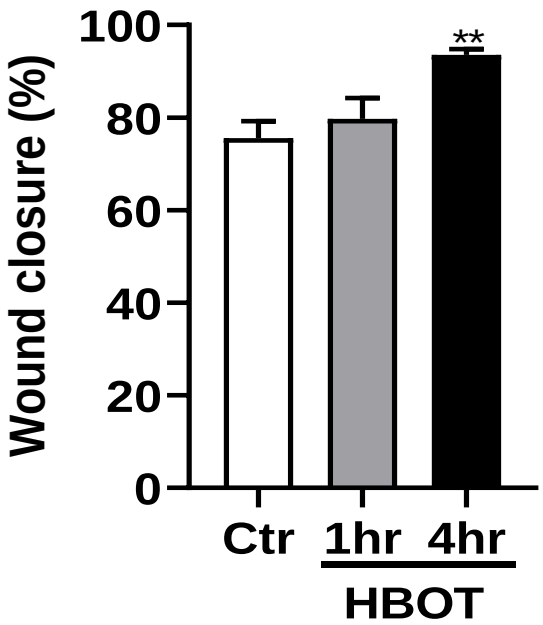
CO 중독 후 TTC (2,3,5-triphenyltetrazolimm chloride) 염색을 통한 뇌손상 부위 확인

동물실험연구

2 창상동물모델에서 고압산소 치료의 치유 효과(wound healing)

- 창상 동물모델에서 고압산소치료(100% 산소, 3기압)에 의한 치유 효과 관찰



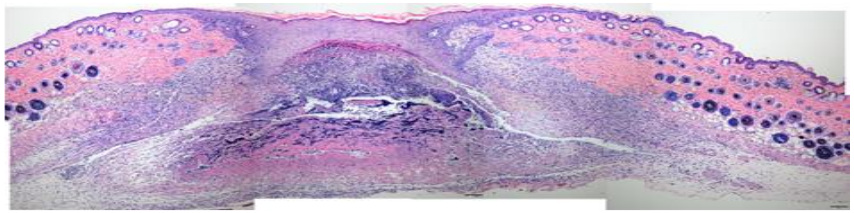


동물실험연구

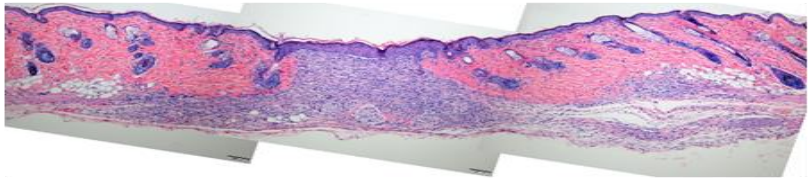
2 창상동물모델에서 고압산소 치료의 치유 효과(wound healing)

- 고압산소치료에 의한 창상 부위의 조직학적 치유·증식 효과 확인

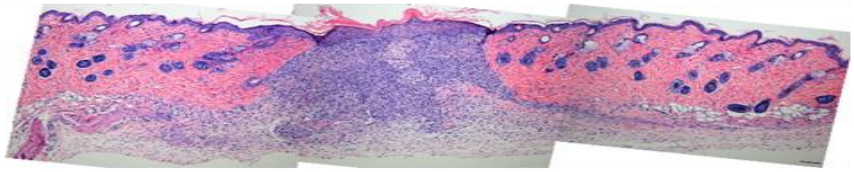
Control



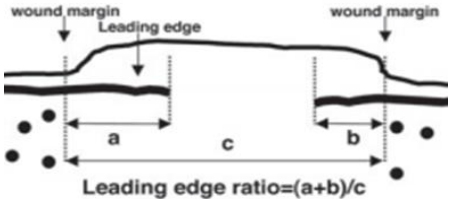
HBOT 1hr

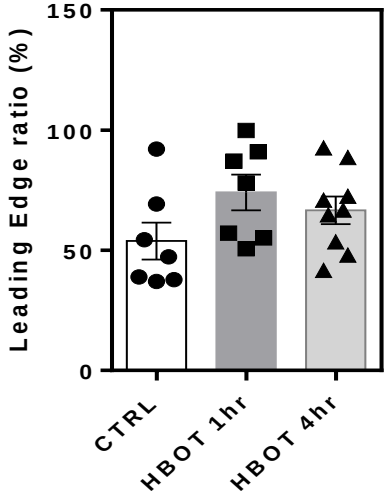


HBOT 4hr



100 μm

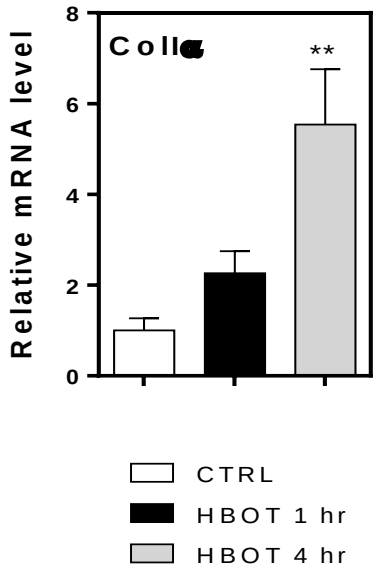


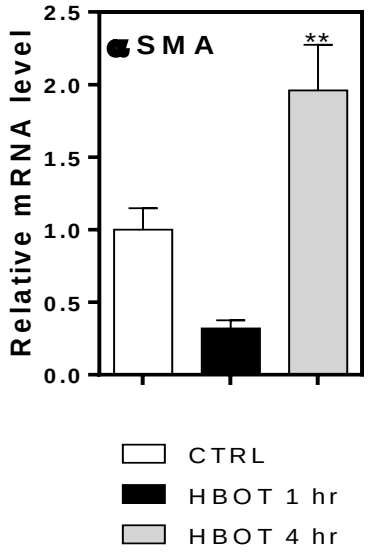


동물실험연구

2 창상동물모델에서 고압산소 치료의 치유 효과(wound healing)

- 고압산소치료에 의한 섬유화 관련 **collagen** 및 **α-smooth muscle actin** 발현 증가





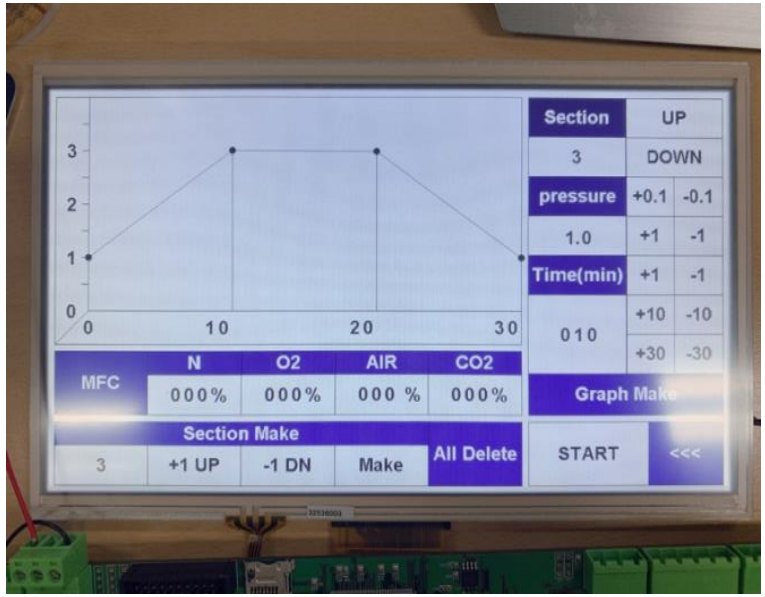
동물실험 추가모델

- Stroke
- MI
- SSNHL
- Etc

세포 연구

3 세포실험을 위한 세포용 고압산소챔버 개발

- **ARM Core(M7)** 기반 세포용 고압산소챔버 제어 **embedded system** 개발 완료
- 세포용 고압산소챔버 외관 및 내부 디자인 설계 완료



세포용 고압산소챔버 **UI**

세포용 고압산소챔버 **Prototype**

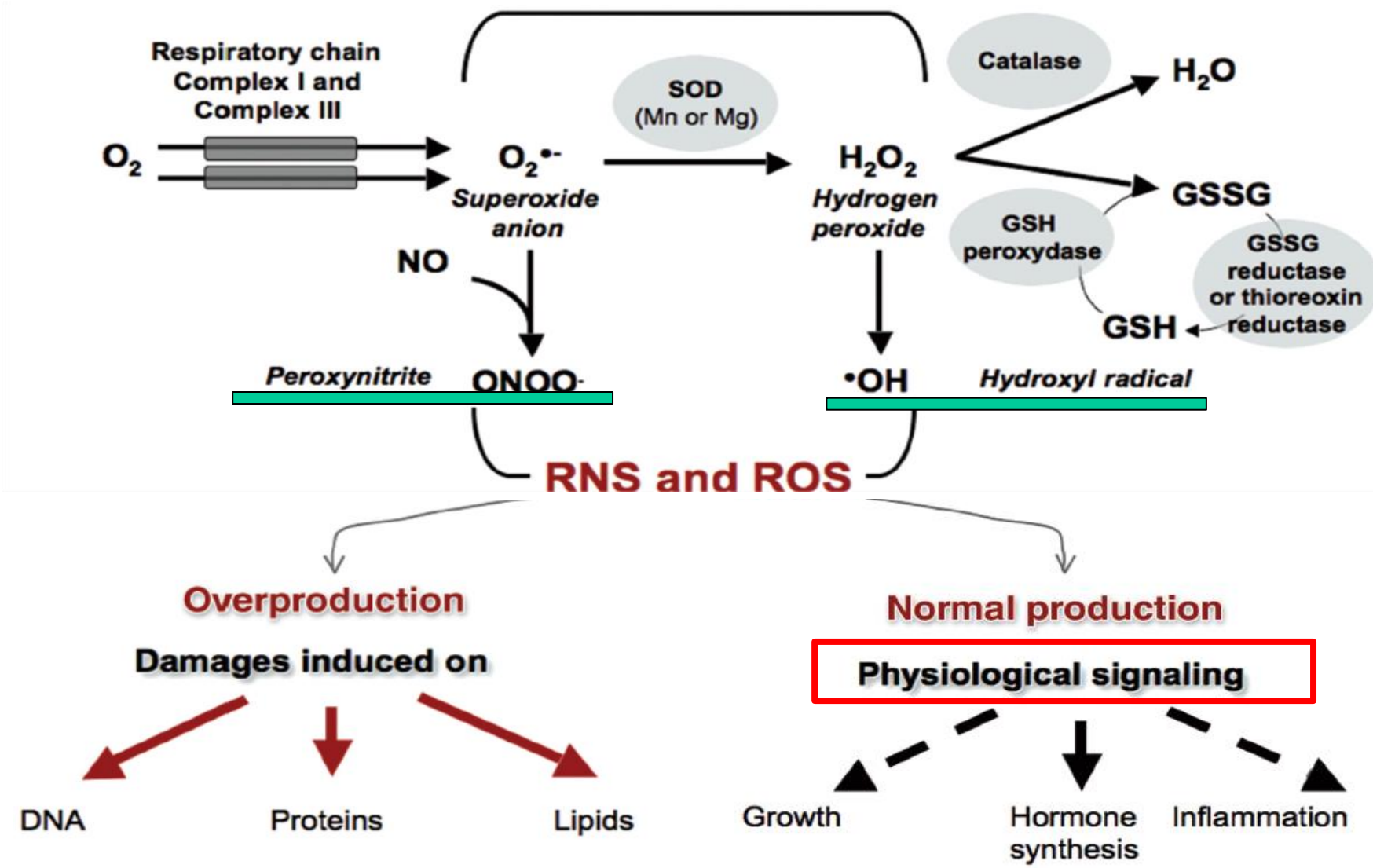
Tertiary mechanism (?)

Mechanisms of HBOT

(Primary, secondary & tertiary)

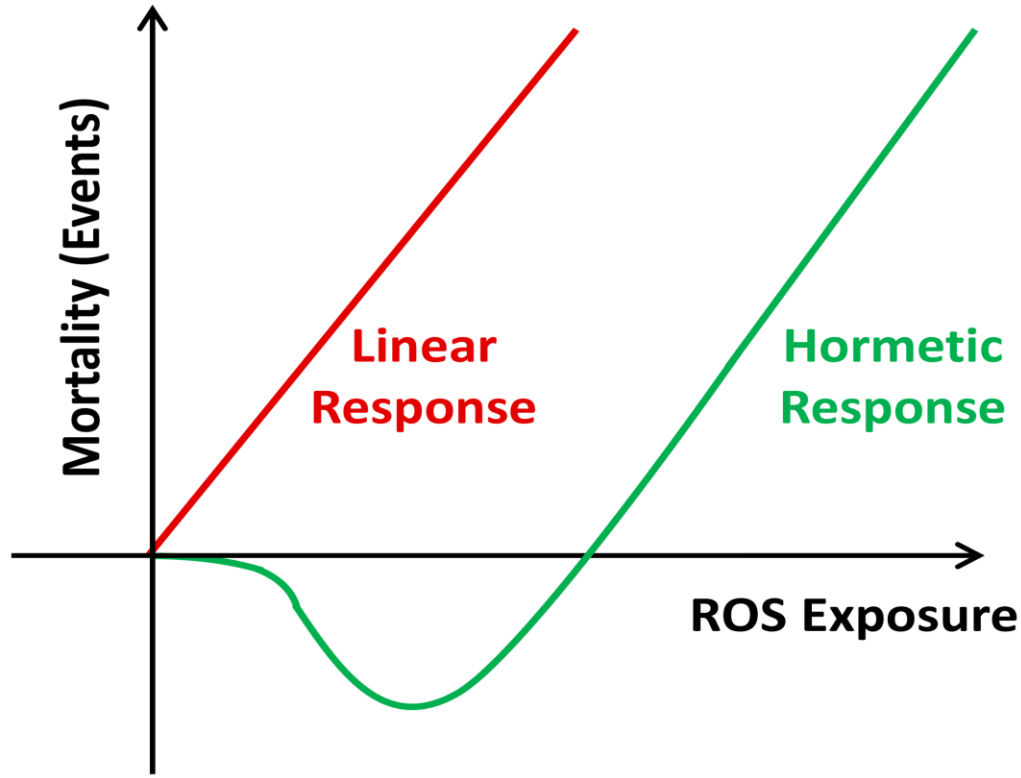
Primary	**Secondary**	**Tertiary**
• Hyperoxygenation (O_2 tensions)	• Enriched O_2 (cellular functions)	• Cellular protection via mitochondria
• Direct effects of		
Oxygen delivery/uptake Bubble reduction	Vascularization Immune response & inflammation Growth factor signaling Induction of antioxidant stress	Mitohormesis ? ? ?

Reactive nitrogen & oxygen



Comprehensive Physiology 2017

Mitochondrial hormesis (mitohormesis)



Free radical theory: linear dose-response relationship (ROS and oxidative stress and mortality)
Mitohormesis: non-linear dose-response relationship (low doses of ROS exposure decrease mortality, while higher doses promote mortality)

Dose Response. 2014 May; 12(2): 288–341

Possible mechanism

Low continuous
or higher intermittent
sub-lethal dose

A coordinated response to
mild mitochondrial stress

Intermittent HBOT

Mitochondrial Stress

Mitohormesis

√ **Reduce oxidative stress**

Most of O_2
in our body
is consumed
in mitochondria

√ **Recover from ischemic damage and promote wound healing**

√ **Resistant to subsequent stress**

Participation in therapeutic mechanism of HBOT

임상 연구

1 정상인의 고압산소 치료에 의한 미토콘드리아 스트레스 바이오마커 변화 측정

- 건강한 정상인 자원자 10명을 대상으로 아래 프로토콜에 따라 고압산소치료 수행 후 일정 시간 간격으로 여러 혈중 지표들의 변화를 측정하였음

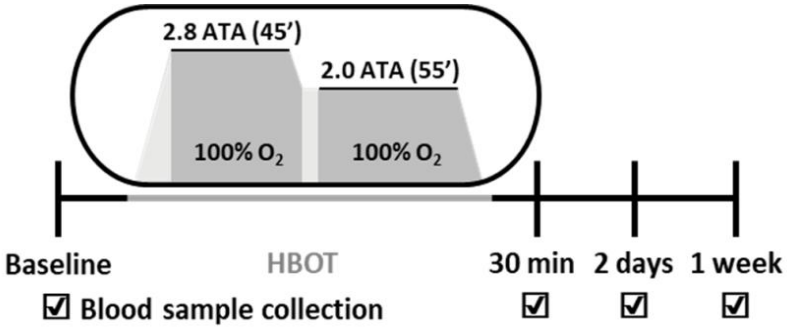


Table 1. Serum biochemical variables at baseline and following acute exposure to hyperbaric oxygen therapy (HBOT).

Variables	Baseline	1 Week Post HBOT	*p*-Value
Glucose (mg/dL)	104.4 ± 19.1	105.0 ± 35.4	0.967
TC (mg/dL)	186.5 ± 17.8	185.8 ± 24.8	0.921
TG (mg/dL) *	154.9 ± 58.4	190.4 ± 66.1	0.012
HDL-C (mg/dL)	47.2 ± 7.5	45.2 ± 7.2	0.206
LDL-C (mg/dL)	121.5 ± 23.1	116.1 ± 23.8	0.272
ALT (U/L) *	39.6 ± 21.2	36.1 ± 12.2	0.483
AST (U/L) *	29.4 ± 5.5	29.7 ± 5.3	0.812
Total protein (g/dL)	7.45 ± 0.26	7.30 ± 0.29	0.143
Albumin (g/dL) *	4.64 ± 0.25	4.58 ± 0.23	0.301
BUN (mg/dL)	12.74 ± 1.65	12.43 ± 1.76	0.599
Creatinine (mg/dL)	0.82 ± 0.05	0.81 ± 0.04	0.532
Uric acid (mg/dL)	6.37 ± 0.85	6.11 ± 0.95	0.106

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BUN, blood urea nitrogen; LDL-C, low density lipoprotein cholesterol; HDL-C, high density lipoprotein cholesterol; TC, total cholesterol; TG, triglyceride. Statistical analyses were performed using Student's paired *t*-test or * Wilcoxon matched-pairs signed-rank test. Values represent the means ± SD.

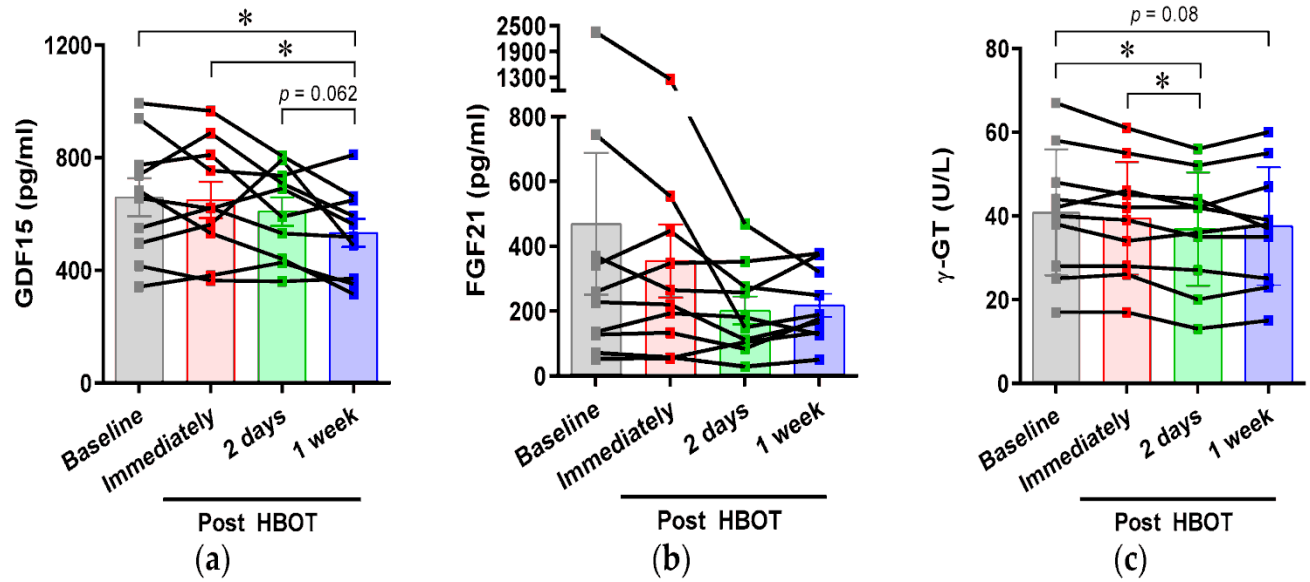
JS Jang, et al. Int. J. Environ. Res. Public Health 2020, 17, 7853

임상 연구

1 정상인의 고압산소 치료에 의한 미토콘드리아 스트레스 바이오마커 변화 측정

- 미토콘드리아 스트레스 바이오마커로 알려진 **GDF15, FGF21, γ-GT** 은 스트레스에 의해 발현 및 분비가 증가하여 이를 경감시키고 회복시키는 작용을 가짐

- 미토콘드리아 스트레스가 높은 상태에서 이들 혈중농도가 증가한다고 알려짐
- 피검자들은 고압산소치료에 의해 **GDF15, FGF21, γ-GT** 혈중 농도가 감소하였으며, 이는 미토콘드리아 스트레스가 줄어들었음을 의미함



(**a**) growth/differentiation factor 15 (GDF15), (**b**) fibroblast growth factor 21 (FGF21), and (**c**) γ-glutamyltransferase (γ-GT)

JS Jang, et al. Int. J. Environ. Res. Public Health 2020, 17, 7853

임상연구

- 각 임상 적응증(16개)에서 mitochondria stress markers
- 새로운 적응증의 개발

챔버연구

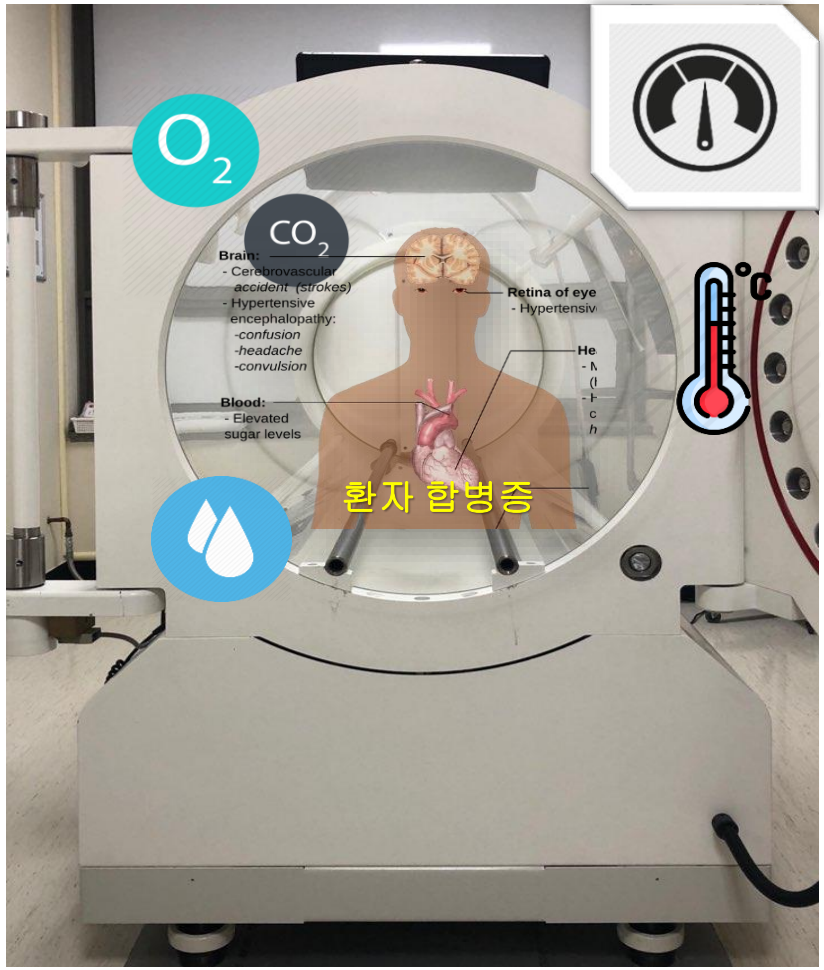
- 정의: 챔버를 이용하여 1.4기압 이상으로 100%산소를 호흡시킴으로 의학적 효과를 나타내는 치료법 (의학적 압력은 2.0 – 3.0 기압)

- 치료하는 동안 압력, 산소등 유지

챔버연구

챔버연구

- 기계적 안정성과 유효성 평가를 위한 외부 측정 장치 사용
- 일인용 고압산소치료기 임상시험 실시
  1. 전향적 데이터(합병증) 수집
  2. 치료 중 고압산소치료기의 압력, 산소, 이산화탄소, 습도 등 측정



CO & CMR (duration)

Methods: CMR studies performed between the years 2005 and 2014 for a primary diagnosis of CO poisoning at a tertiary care center were reviewed by an experienced cardiologist. Variables of interest were compared between patients with normal and abnormal studies to identify factors associated with cardiac dysfunction.

Results: Eighty-eight patients underwent stress CMR, age 34 years (range 11-70); 49% were male, 74 had acute poisoning and 14 had chronic poisoning (CO exposure for longer than 24 hours). Time from CO poisoning to imaging was 24 months (1day-120 months). Patients were stratified into four categories, which included those with acute poisoning imaged:

- ≤12 months;
- 12-60 months;
- >60 months from the event; and

Table 1. Patient characteristics and detail of stress CMR abnormalities according to time interval from poisoning to imaging

	Category by time interval from incident to imaging			
	≤12 mos.	13-60 mos.	>60 mos.	chronic poisoning
N*	26	32	12	14
age	34 (17-69)	35 (11-58)	33 (19-61)	33 (28-70)
male sex	14 (54%)	14 (42%)	8 (67%)	5 (38%)
HTN	1 (4%)	2 (6%)	3 (35%)	1 (7%)
CAD	0	1 (3%)	0	1 (7%)
HLD	0	1 (3%)	2 (17%)	1 (7%)
active smokers	7 (27%)	2 (6%)	5 (42%)	10 (71%)
troponin I – ng/dl (IQR); N=22	0.27 (0.03, 3.2)	0 (0, 3.6)	0 (0, 1.27)	0.31 (0, 0.63)
COHb -% (IQR)	15 (11.5, 19.8)	18.4 (11.7, 22.2)	12 (9.7, 18.7)	--
loss of consciousness	8 (31%)	8 (25%)	7 (58%)	3 (21%)
LV dysfunction†	2 (8%)	8 (26%)	2 (17%)	2 (17%)
LVEF - (% normal vs abnormal)	63 (56-79) vs. 53 (51-54)	67 (56-85) vs. 50 (42-53)	65 (56-78) vs. 52 (49-54)	68 (58-73) vs. 52 (51-52)
LV dilatation	2 (8%)	1 (3%)	2 (17%)	1 (8 %)
LVEDVI (mL/m^2)	70 (35-91) vs. 97 (95-100)	75 (55-90) vs. 122	78 (54-89) vs. 95 (94-96)	66 (52-73) vs. 120
RV dysfunction	5 (20%)	3 (9%)	0	1 (9%)
ischemia	1 (4%)	0	1 (8%)	0
scar	0	0	1 (8%)	0

Miguel Alvarez Villela, UHM 2020, VOL. 47

CO & CMR (duration)

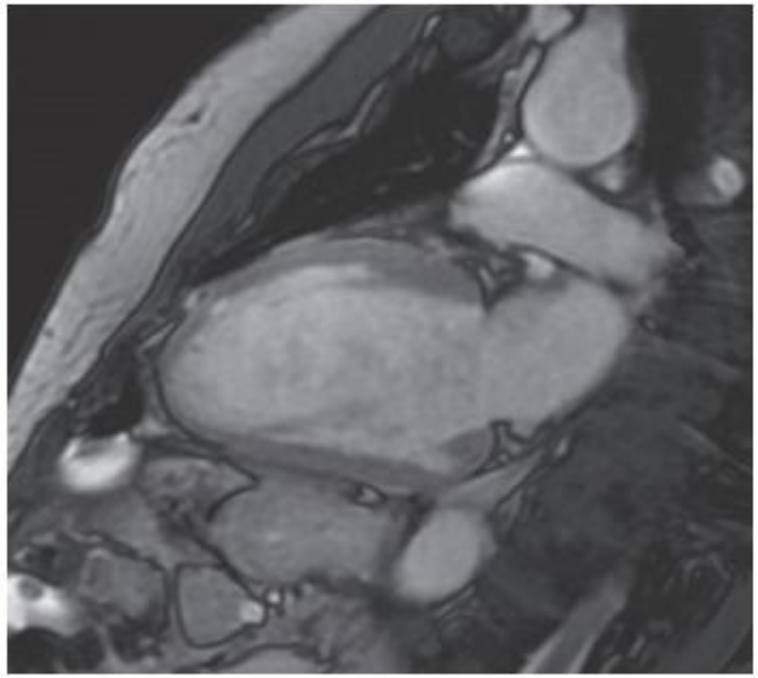


Figure 2. Remodeled left ventricle in a patient with acute CO poisoning. Dilated, spherically remodeled left ventricle (two-chamber view, end-diastole). Seen in 23% of the abnormal studies after CO poisoning.

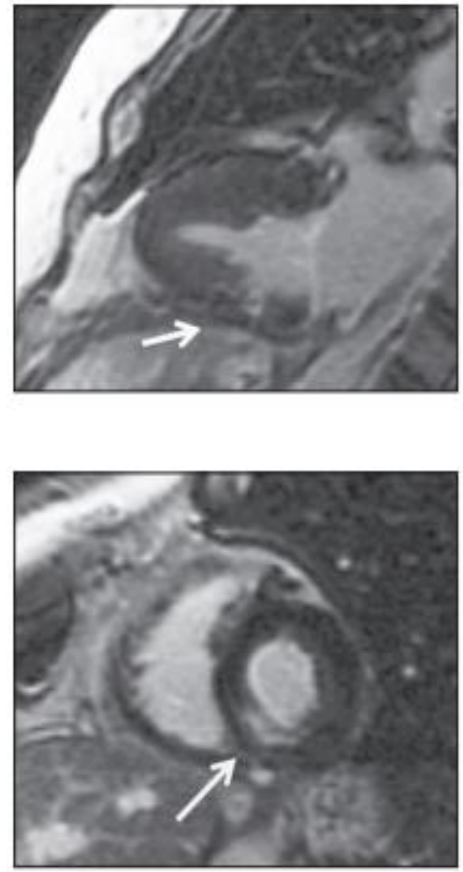


Figure 3. Myocardial scarring in a patient with chronic CO poisoning.

Short-axis and two-chamber late gadolinium enhancement sequences showing a small inferior/inferoseptal scar. Seen in only one patient after chronic CO exposure.

Miguel Alvarez Villela, UHM 2020, VOL. 47

앞으로의 연구

Indications (16)

- **Air or Gas Embolism**
- **Arterial Insufficiencies**
- **Carbon Monoxide Poisoning**
- **Clostridial Myonecrosis**
- **Compromised Grafts and Flaps**
- **Crush Injuries and Skeletal Muscle**
- **Decompression Sickness**
- **Delayed Radiation Injuries**
- **Idiopathic Sudden Sensorineural Hearing Loss**
- **Intracranial Abscess**
- **Necrotizing Soft Tissue Infections**
- **Refractory Osteomyelitis**
- **Severe Anemia**
- **Thermal Burns**
- **Diabetic Foot Ulcer**
- **Central Retinal Artery Occlusion**

Indications

- Emergency

① **Decompression Sickness, air or gas embolism (AGE)**
② **CO, severe anemia**
③ **Arterial Insufficiencies, CRAO, ISNHL**

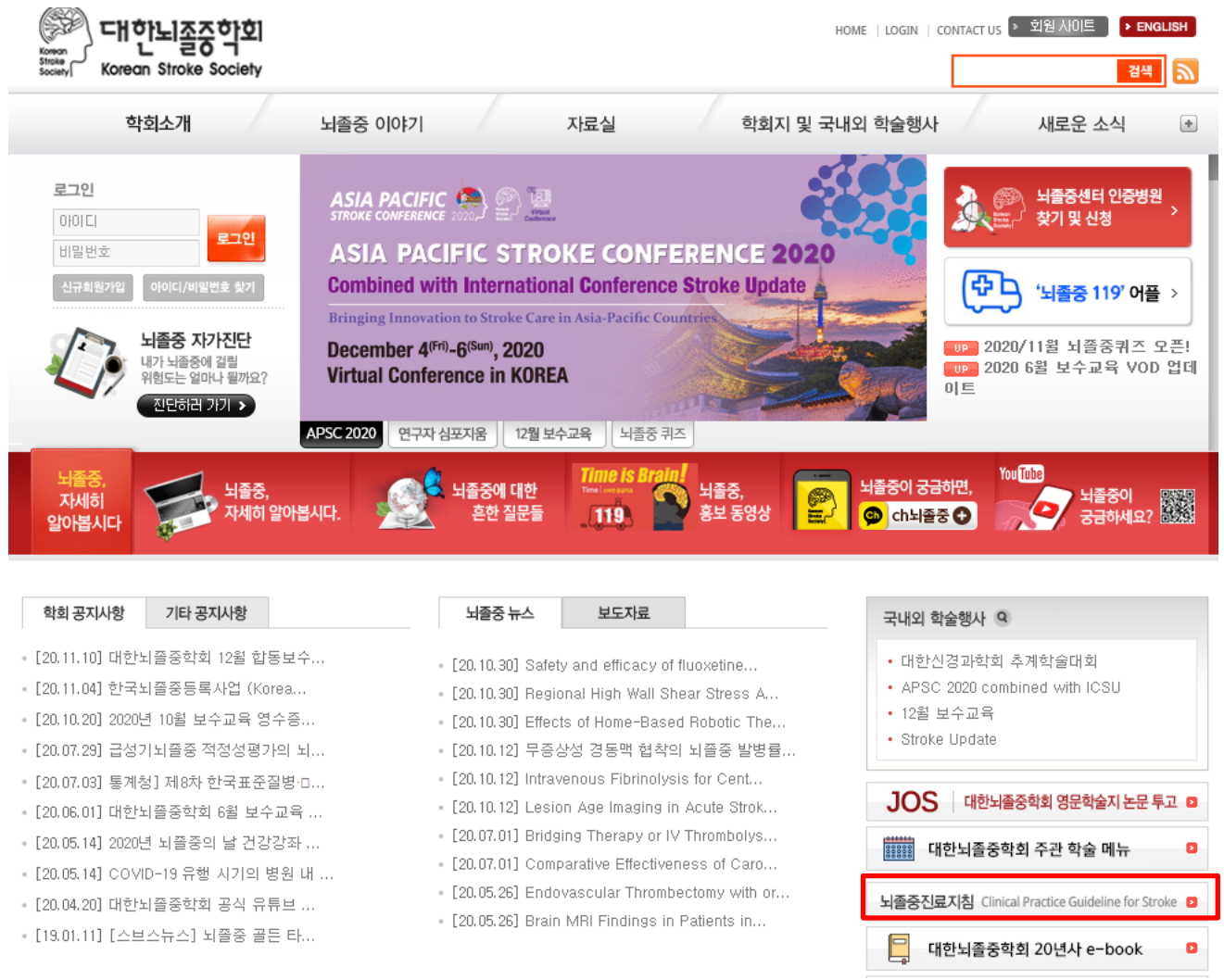
- Elective: 4 categories (wounds)

① **Chronic**: refractory diabetic wounds, arterial insufficiency ulcers
② **Radiation-induced**: delayed radiation injury, radionecrosis
③ **Infected**: gas gangrene, osteomyelitis, necrotizing soft tissue infection, brain abscess
④ **Traumatic**: crush injury, compromised skin grafts and flaps, thermal burn

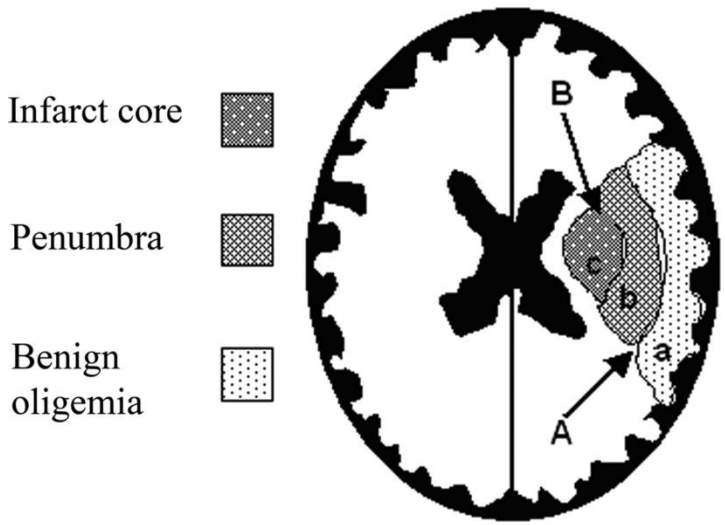
Acute stroke

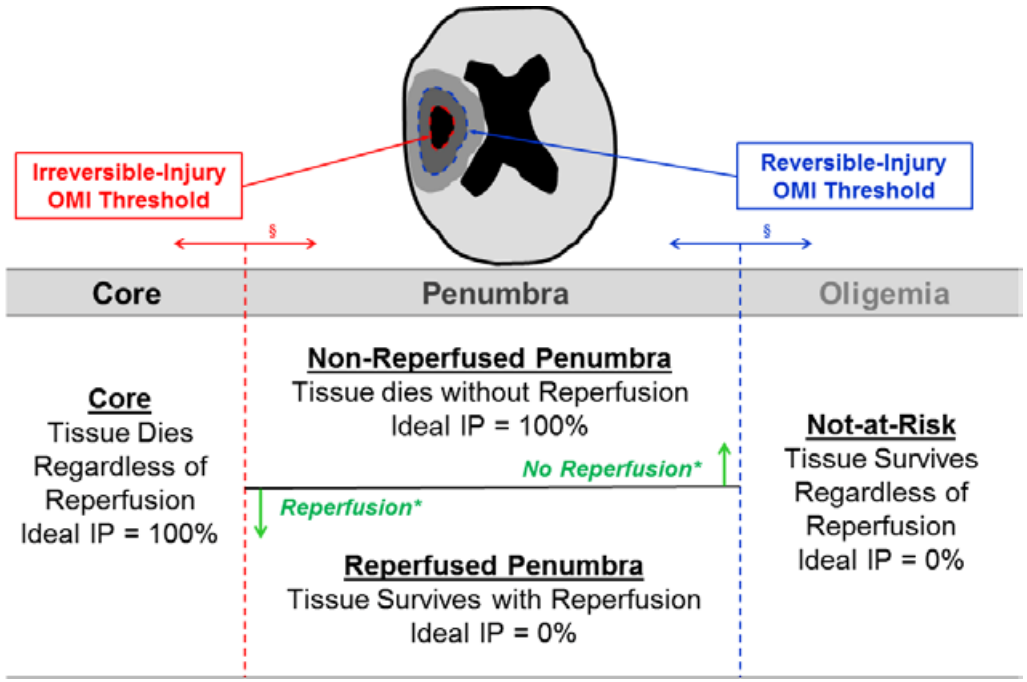
- Time criteria – time is brain
- 3 hr, 4.5 hr, 6 hr ….
- DAWN, DEFUSE-3 trials -> 16 hr, 24 hr

Acute stroke



Acute stroke





Acute stroke

2.3.3. 혈전용해술 Thrombolysis

2.3.3.1. 정맥내 혈전용해술 Intravenous Thrombolysis

권고사항
1. 허혈성 뇌졸중 환자에서 증상 발생 (최종적으로 정상이었던 시간 기준) 3시간 이내에 투여가 가능 한 경우, IV tPA 치료를 한다. (근거수준 Ia, 권고수준 A) 단, 영상검사에서 뇌출혈이 배제되어야 하며, 그외 적응증은 표 2. 에 따른다. - 문구 개정
2. 위의 치료는 81세 이상의 환자에서도 적용 가능하다. (근거수준 Ib, 권고수준 A) - 추가
3. 허혈성 뇌졸중 환자에서 증상 발생 (최종적으로 정상이었던 시간 기준) 3~4.5 시간 사이에 투여가 가능한 경우 IV tPA 투여를 할 수 있다. (근거수준 Ia, 권고수준 A) - 추가 * 단, 80세 이상, 심한 신경학적 장애 (NIHSS점수 25점 초과), 과거 뇌졸중과 당뇨병의 병력이 함께 있는 환자, 경구 항응고제를 복용하고 있는 환자에서는 3~4.5 시간 투여할 근거가 부족하다.

Recommendations

Endovascular Recanalization Therapy (ERT)

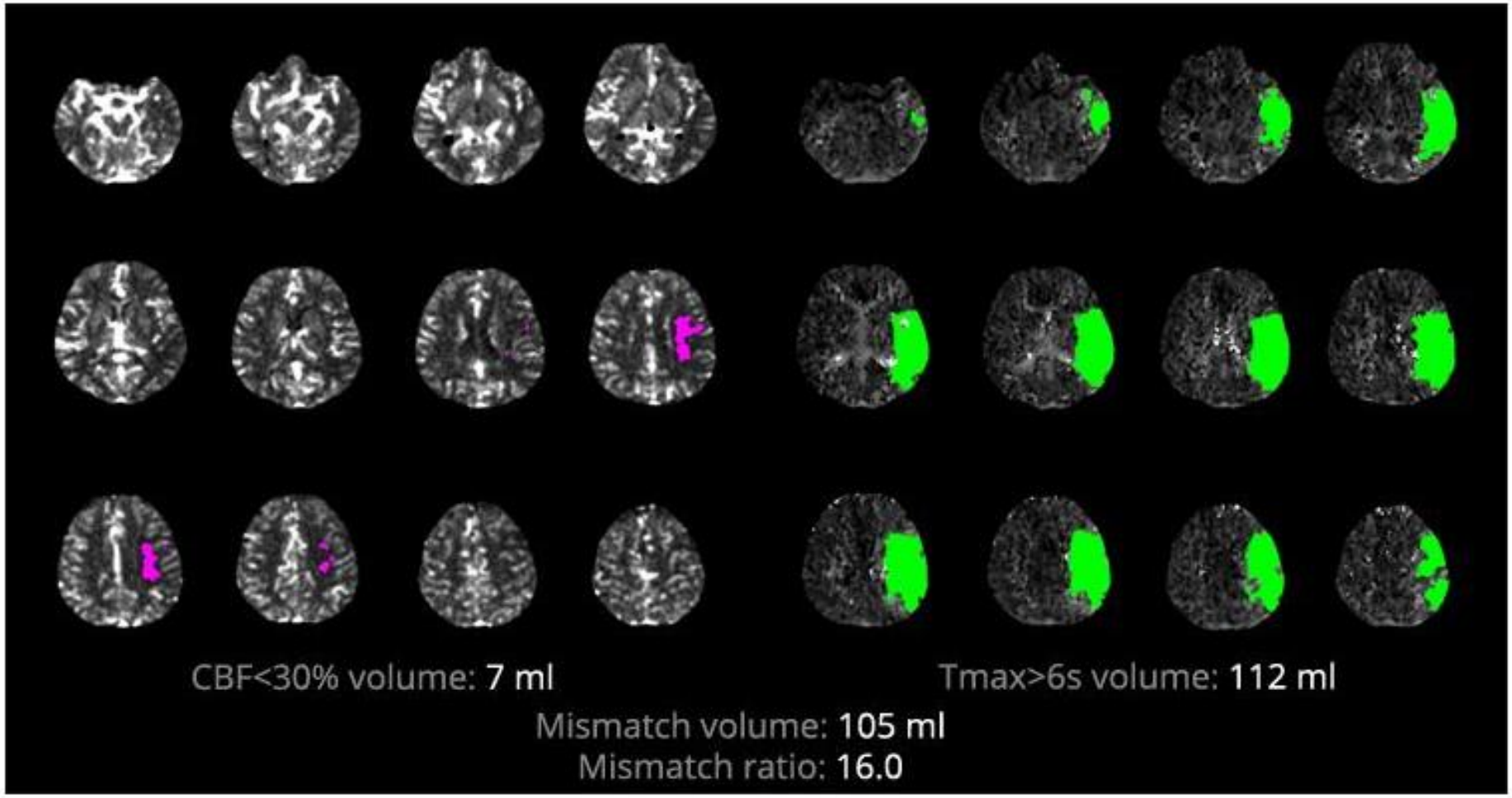
1. In patients with major ischemic stroke due to an acute large artery occlusion in anterior circulation (internal carotid artery, M1, and possibly large M2 branch) within 6 hours, ERT is recommended to improve clinical outcomes (level of evidence [LOE] Ia, grade of recommendation [GOR] A).
2. In patients eligible for intravenous tissue plasminogen activator (IV-TPA), administration of IV-TPA is recommended before the initiation of ERT (LOE Ia, GOR A). Since IV-TPA should not significantly delay ERT, it is recommended to simultaneously proceed ERT during IV-TPA treatment without waiting for clinical response to IV-TPA.
3. In patients who are contraindicated for IV-TPA, ERT is recommended as a first-line therapy in patients with major ischemic stroke due to an acute large artery occlusion in the anterior circulation within 6 hours (LOE IIa, GOR B).
4. In patients with major ischemic stroke due to acute large artery occlusion in the poster circulation (basilar artery, P1, and vertebral artery) within 6 hours, ERT can be considered (LOE III, GOR B).
5. In selected patients with acute ischemic stroke due to large vessel occlusion in the anterior circulation presenting within 6 to 24 hours from last seen normal, endovascular recanalization therapy can be recommended when the patients have target mismatch assessed by multimodal imaging and/or clinical deficit AND when reperfusion by ERT is expected to improve the outcome of the patients. For patient selection, each institution is recommended to have its own criteria, which can timely and reasonably identify patients with target mismatch in the late time window (LOE III, GOR C).

Acute stroke

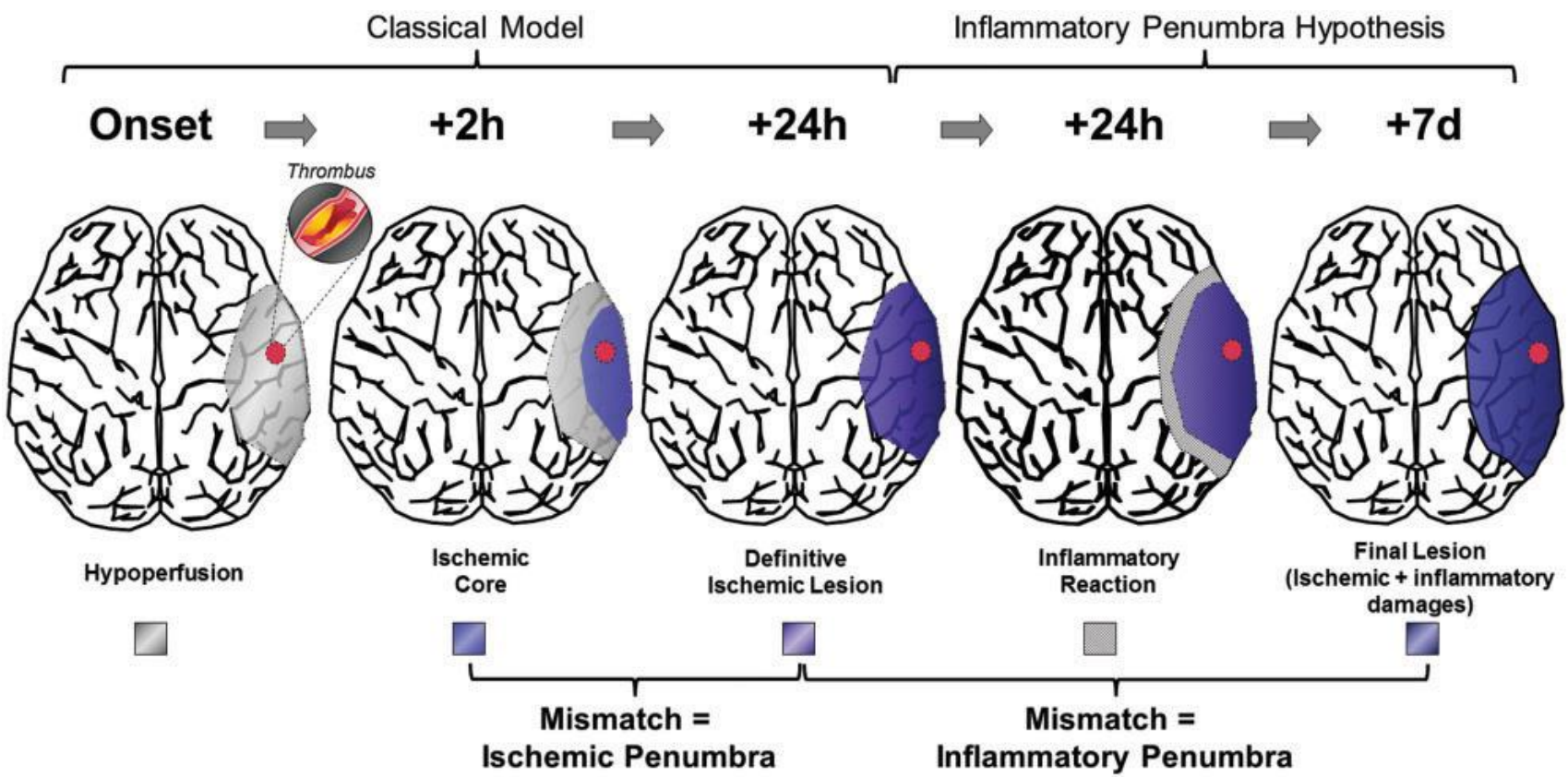
Imaging Criteria

1. **ICA or MCA-M1 occlusion** (carotid occlusions can be cervical or intracranial; with or without tandem MCA lesions) by MRA or CTA
2. **Target Mismatch Profile** on CT perfusion or MRI (ischemic core volume is <70 ml, mismatch ratio is ≥1.8 and mismatch volume is ≥15 ml)

Acute stroke

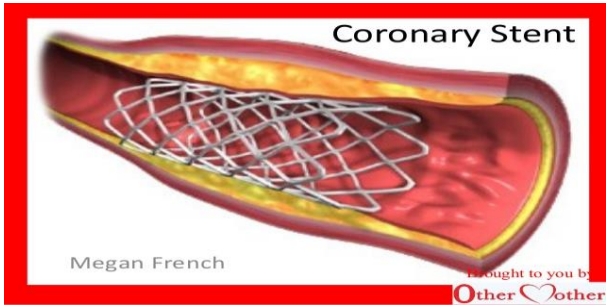


The inflammatory penumbra concept after ischemic stroke



European Stroke Journal, 2016, Vol. 1(1) 20–27

Vascular emergency



Macrocirculation

Vascular intervention

Microcirculation

HBOT

AMI

- Time criteria – time is muscle
- 12 hr ….
- Trials -> 12 – 24 hr (pain, shock, arrhythmia)

AMI

대한심장학회
THE KOREAN SOCIETY OF CARDIOLOGY

진료지침

HOME | 진료지침 | 진료지침

전체

제목/내용

[2020년/국내] 수술 및 시술 시 항혈전제 사용에 대한 전문가 합의문
2020-11-16

[2020년/국내] 급성심근경색증의 약물치료에 대한 전문가 합의문
2020-10-22

[2020년/ESC] ESC Guidelines for the diagnosis and management of atrial fibrillation developed in collaboration with the EACTS
2020-09-18

[2020년/ESC] ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation
2020-09-18

[2020년/ESC] 2020 ESC Guidelines on Sports Cardiology and Exercise in Patients with Cardiovascular Disease
2020-09-18

[2020년/ESC] 2020 Adult Congenital Heart Disease (previously Grown-Up Congenital Heart Disease) (Management of) Guidelines
2020-09-18

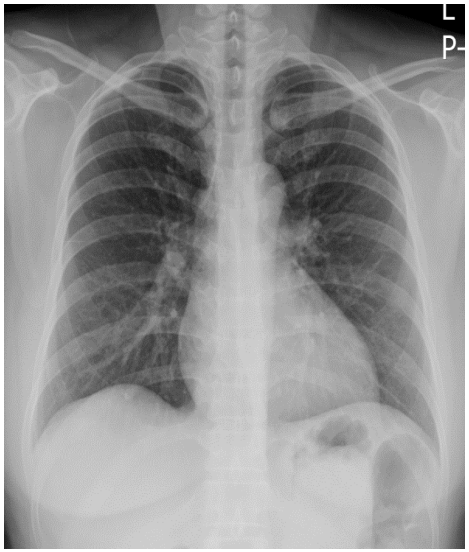
[2020년/국내] 코로나이러스 감염증-19에 대한 대한내과학회 진료 지침
2020-05-18

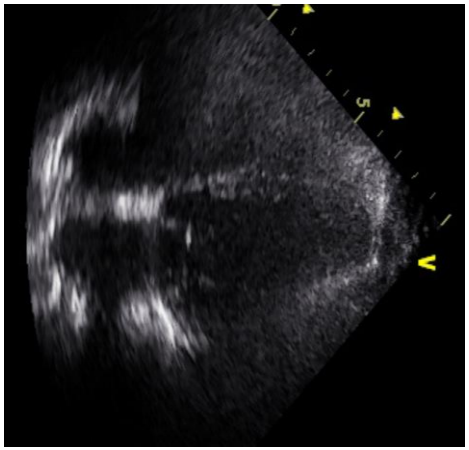
[2020년/국내] 급성 심부전 진료지침 2020 업데이트
2020-03-16

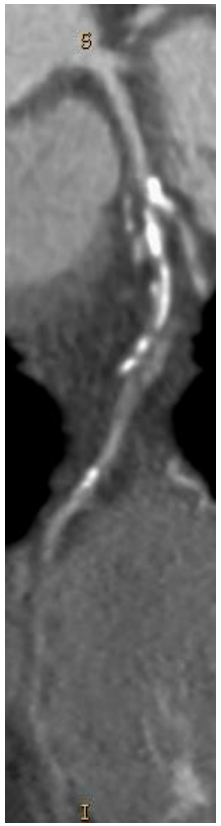
[2019년/ACC/AHA] 2019 AHA/ACC/HRS Focused Updateof the 2014 AHA/ACC/HRS Guidelinefor the Management of Patients WithAtrial Fibrillation
2019-09-27

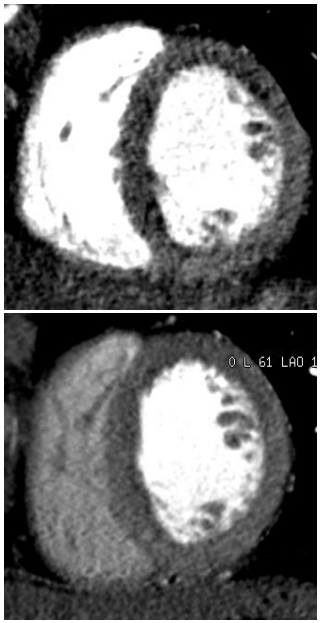
AMI (intervention + drug + ?)

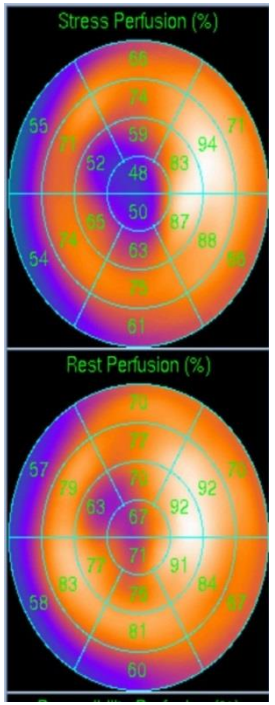
49/F
chest pain and dyspnea
DM, HTN, smoking, hyperlipidemia

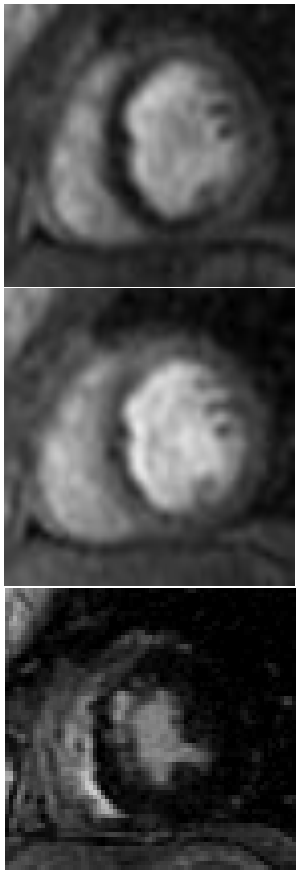








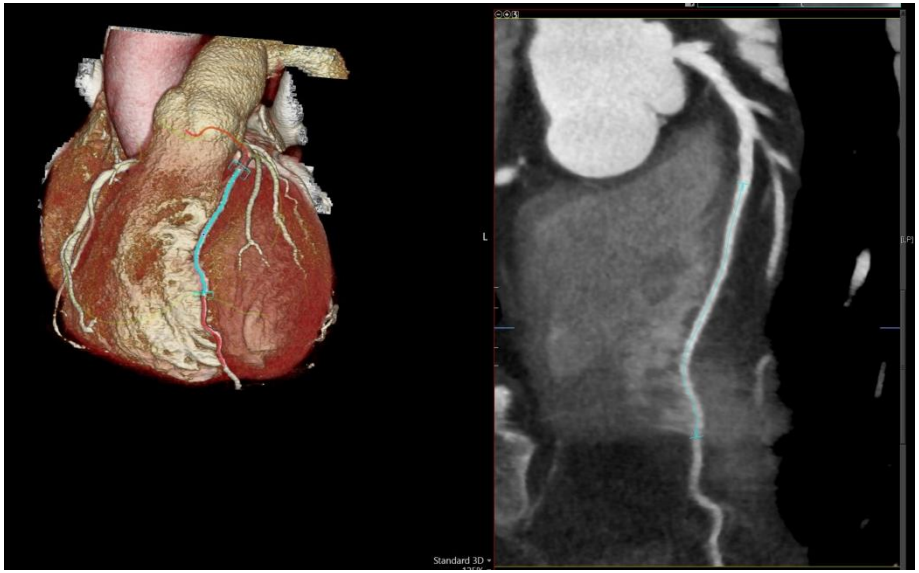




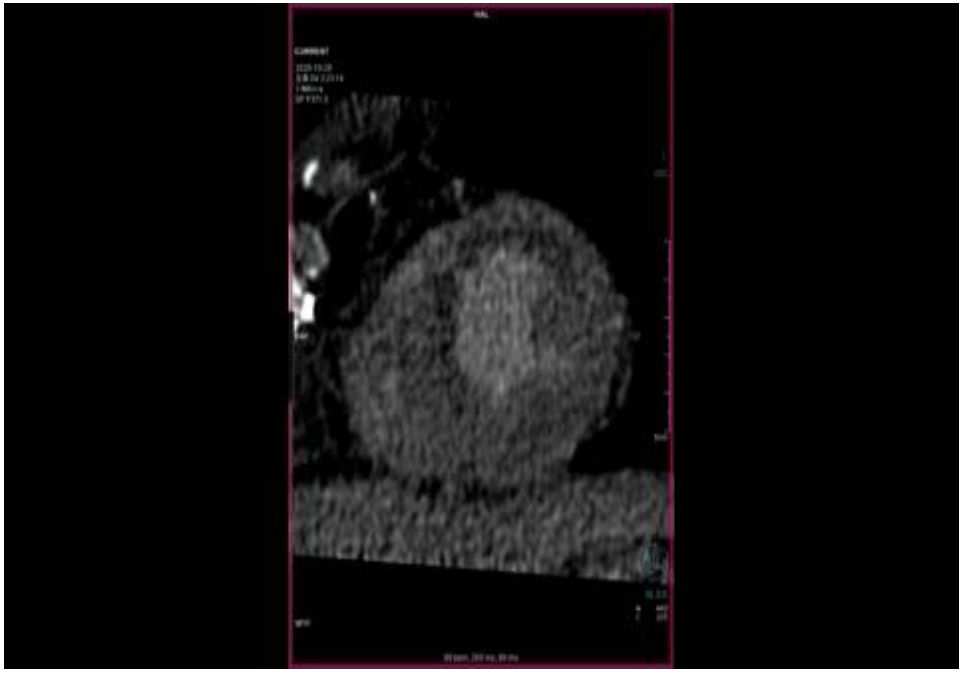
Stress perfusion CT *SPECT* *CMR*

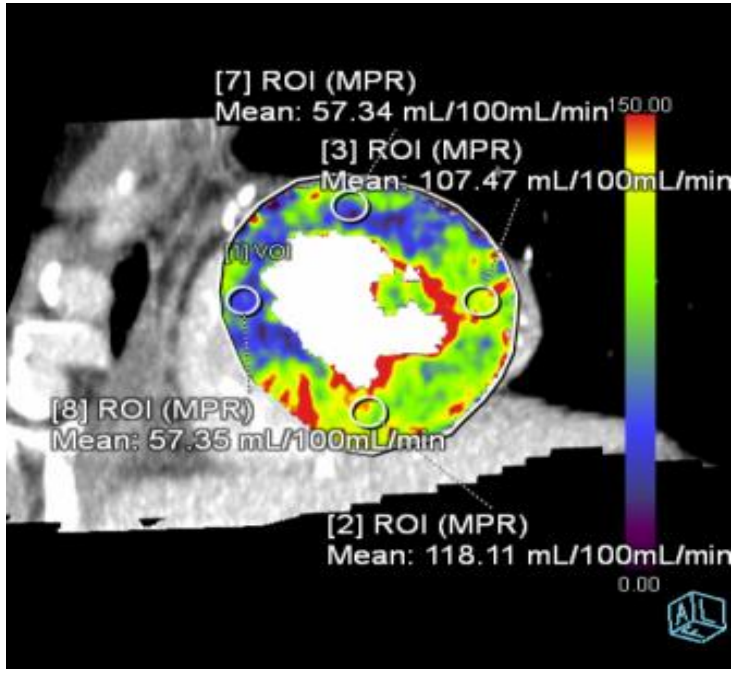
KO SM et al. Int J Cardiovasc Imaging

Large vessel: intact
Treatment: drug + ?

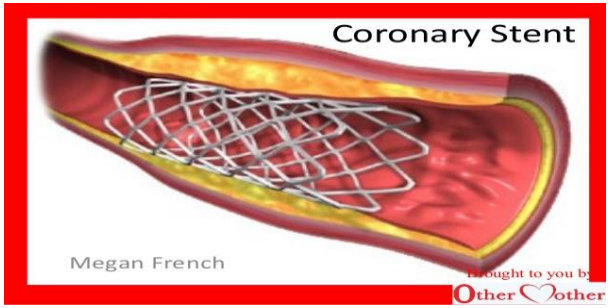


55/M
Cardiac enzyme (-), echo (LVH, no RWMA)





Vascular emergency



Macrocirculation

Vascular intervention

Microcirculation

HBOT

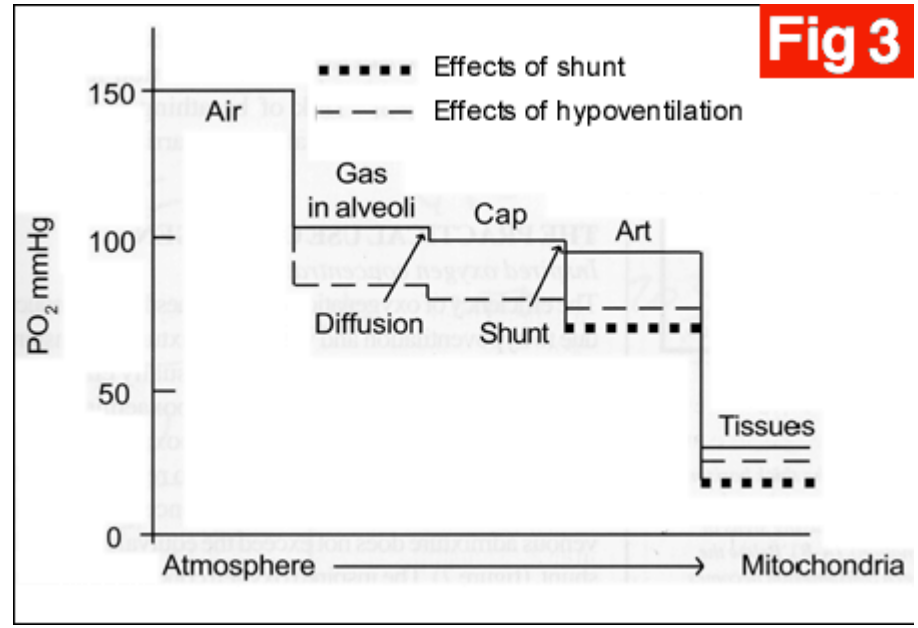
Study design

- AMI, stroke animal model: evidence
- AMI
  - Conventional group
  - Study group (HBO_2)

Clinical & imaging (MRI) study

Movement of O_2 down

- Oxygen concentration: down (air -> cell)
- PO_2: the lowest level (4-20 mmHg)
  in the mitochondria



Life without blood

Dr. Ite Boerema

Life without blood

(A study of the influence of high atmospheric pressure and hypothermia on dilution of the blood)

by

I. BOEREMA (*), N. G. MEYNE, W. K. BRUMMELKAMP
S. BOUMA, M. H. MENSCH, F. KAMERMANS, M. STERN HANF
and W. VAN AALDEREN
(from the Surgical Department of the University of Amsterdam)

When in 1948 we (first at research) started our experiment on hypothermia[1] [2] our ultimate aim was to reduce the metabolism of a warm-blooded animal to such an extent that all the physiological processes would almost come to a standstill.

If successful, this would enable the heart to be clamped off for a period long enough to allow for a major intracardiac operation to take place. When, however, we presented our results to the Netherlands Society of Surgeons in 1950 this aim had not been achieved by any means. In a hypothermic animal at about 27° C., the circulation could be stopped with good chances of survival for about twice as long as in a normothermic animal. The gain in time, about 100 per cent, was relatively great, but absolutely it was very modest, amounting to about five minutes; the reason for this was that below 20° C the physiology was altered too much and the normal harmony of life processes disturbed too much to allow for continuation of life or normal recovery by warming up.

Efforts to achieve safe conditions at a lower level of hypothermia so as to gain a greater period of time for clamping off the heart failed until recently, at any rate for animals with the same weight as human patients. So in 1956 we presented a series of experiments which showed that it was possible to clamp off the circulation for a greater length of time without lowering the temperature further than 27° C.[3] [4] We operated on the animal in a pressure chamber at an absolute pressure of three atmospheres. The animal breathed pure oxygen, the investigators naturally breathed air.

Through the combination of inhaling pure oxygen and being under three atmospheres of pressure, the whole body was supersaturated with oxygen in physical solution.

(*) Professor of Surgery.

Again we are much indebted to T.N.O. (Institute for Applied Physical Research) for financial aid, to the staff of the Mijnendienst of the Royal Dutch Navy at Den Helder and to our advisor on technical matters, P. Bouma.

J Cardiovasc Surg 1959;13:133-146

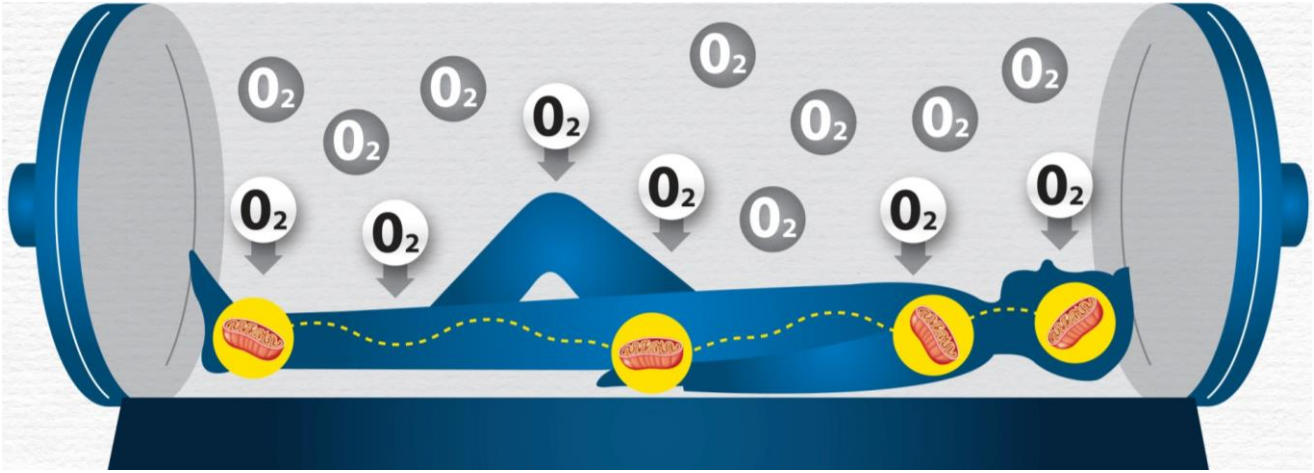
Life is in the blood

- Present: large vessel (Hb) - intervention

$CaO_2 = \boxed{1.34 \times Hb \times SaO_2} + \boxed{0.003 \times PaO_2}$

Hg-bound | Dissolved

- Future: large vessel + small vessel – intervention + HBO_2



I have a dream

HICU

Stroke, AMI, vascular emergency