파크병원
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Hyperbaric oxygen therapy Stimulate to Natural Killer cell activity in cancer patients

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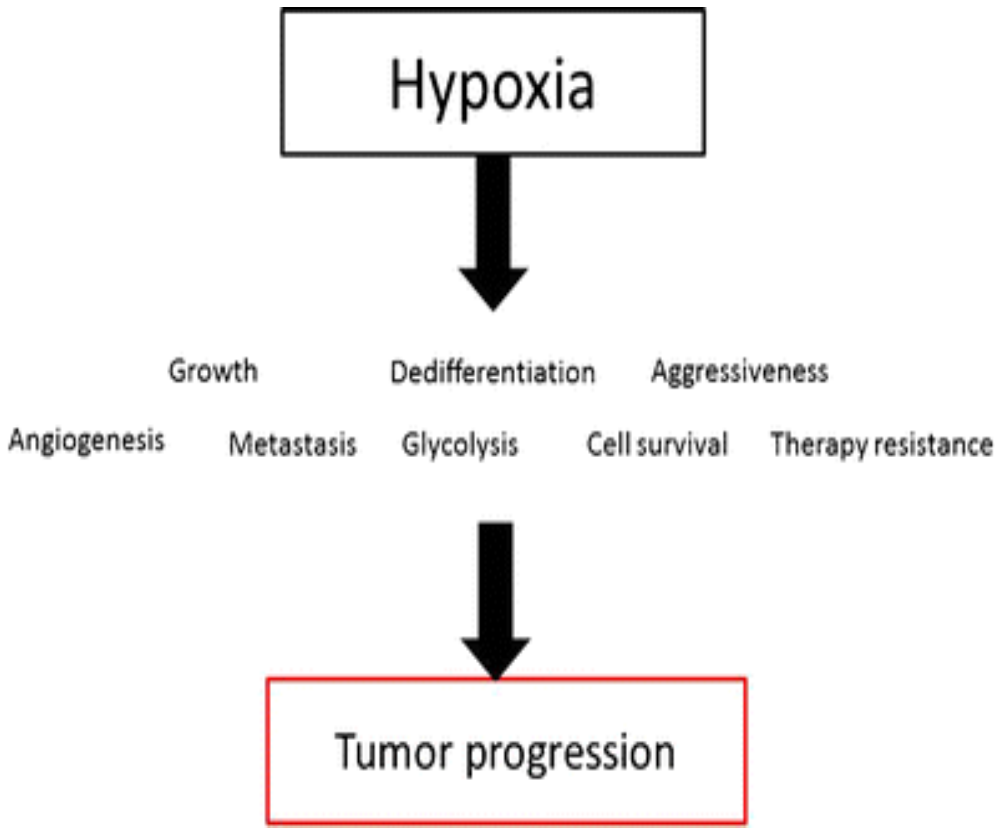
Hypoxia on tumors



- Hypoxia is reported to result in cellular responses which improve oxygenation and viability through induction of angiogenesis, an alteration in metabolism by increased glycolysis and upregulation of genes involved in cell survival/apoptosis
- Hypoxia is a critical hallmark of solid tumors and involves enhanced cell survival, angiogenesis, glycolytic metabolism, and metastasis.

I Moen et al. *Targ Oncol* **7,** 233–242 (2012)

Hyperbaric oxygen(HBO) therapy and cancer

- Hyperbaric oxygen can be used to overcome hypoxia.
- As in normal tissue, the pO2 in cancer tissue increases significantly during HBO exposure.
- Elevation of the tumor oxygen pressure has been shown to be preserved clinically for approximately 30 min after HBO exposure.

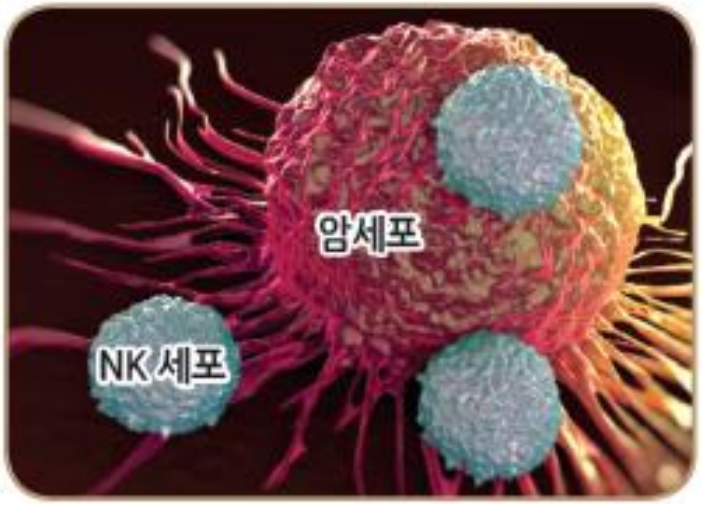
I Moen et al. *Targ Oncol* **7,** 233–242 (2012).

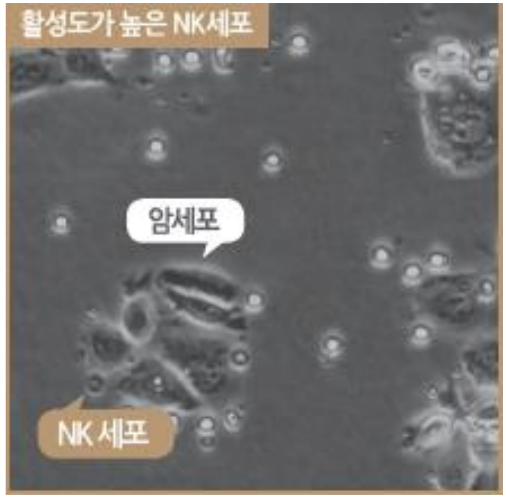
Hyperbaric oxygen therapy and cancer

- Hypoxia has proven to be a causative factor in many pathophysiological events, including cancer progression.
- Decreased oxygen tension selects for more malignant cells and induces multiple cellular adaptations, which again sustains cancer progression and cancer growth.
- Today, angiogenesis is proposed to be a key factor for cancer growth and metastasis.

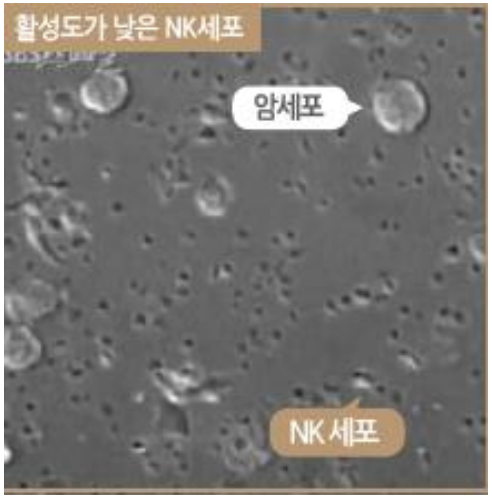
UHM 2003, vol.30 No.1 -hyperbaric oxygen and cancer growth
*I Moen et al. Targ Oncol **7**, 233–242 (2012).*

NK cell activity vs cancer cells



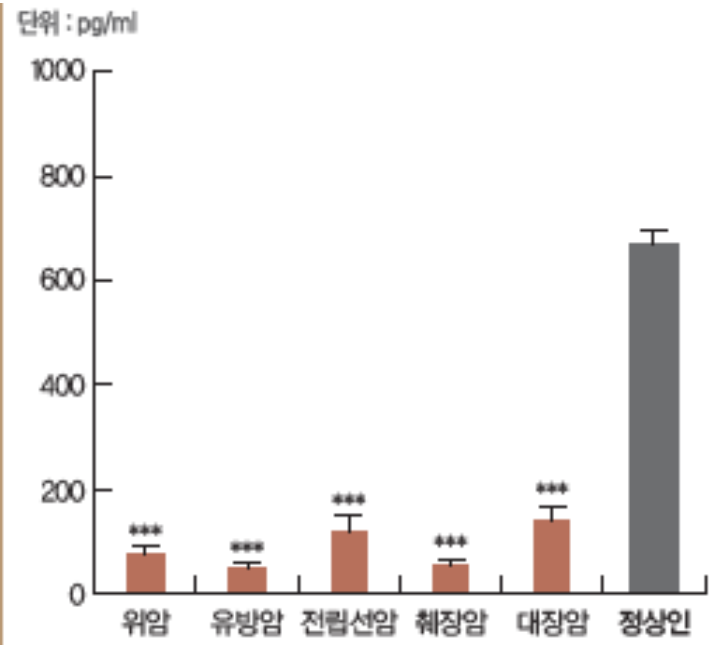


활성도가 높은 NK세포

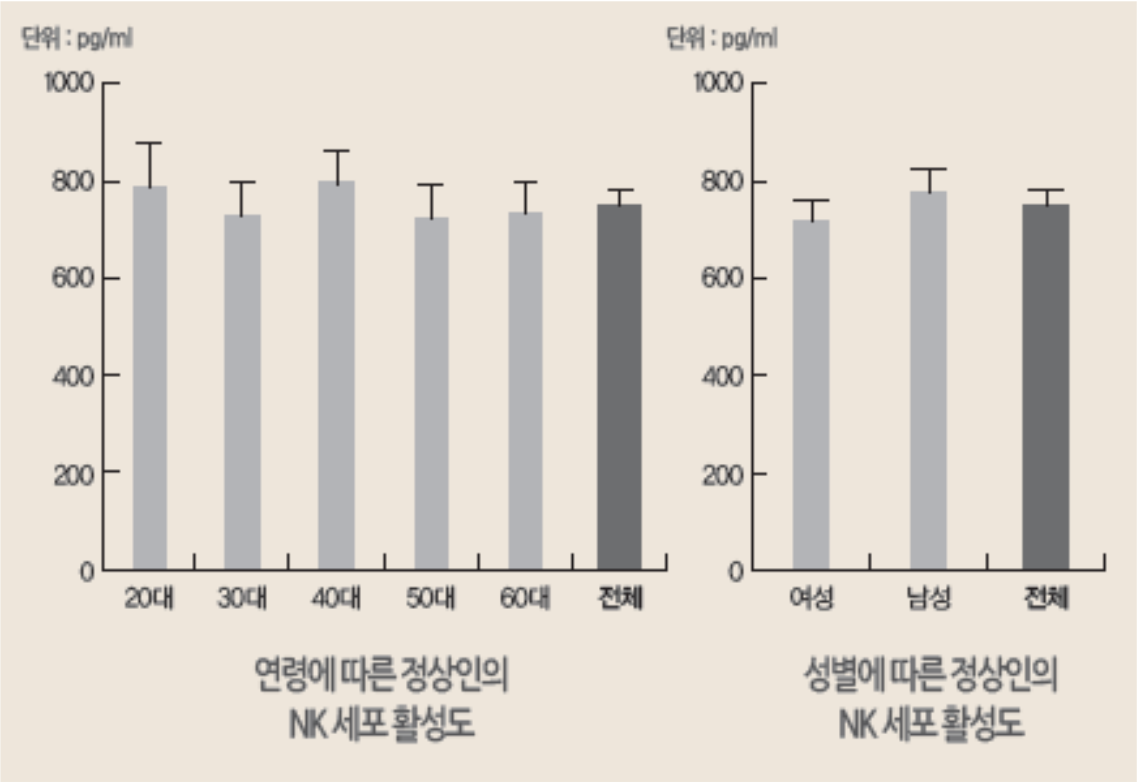


활성도가 낮은 NK세포

NK cell activity vs cancer cells

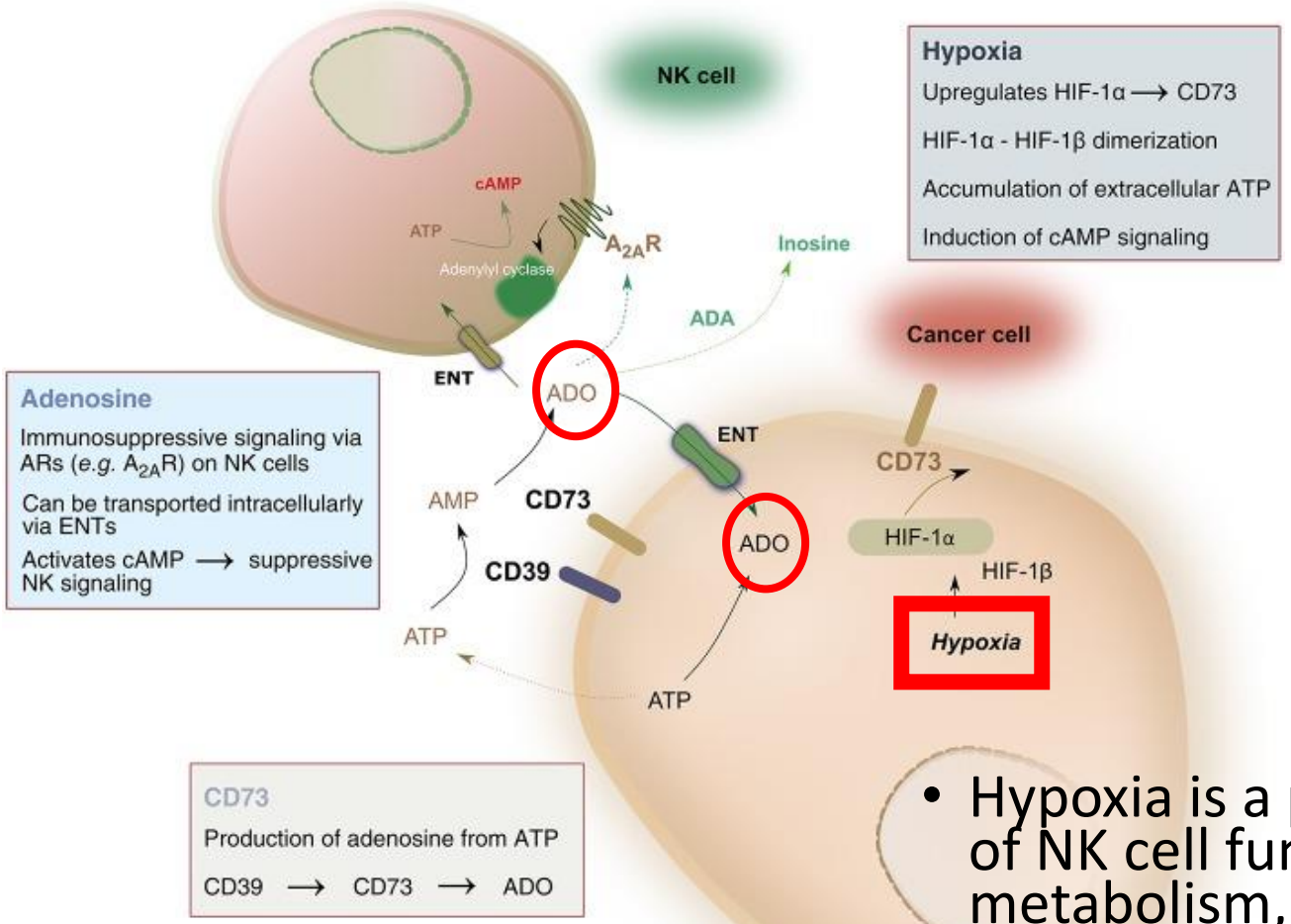


5대 암 환자의 NK 세포 활성도



<임상시험 : 연세대 세브란스병원 임상시험센터>

Hypoxia-CD73 signaling on NK cells



- Hypoxia is a powerful mediator of NK cell function and metabolism, as well as tumor progression.

Chambers AM, Matosevic S. *Front Mol Biosci*. 2019;6:60

Effects of hypoxia on NK cell function and metabolism

TABLE 1 | Effects of hypoxia on NK cell function and metabolism.

Immunomodulatory factor	Effect	References
Oxygen level		
Hypoxia	Downregulation of activating NK receptors NKp46, NKp30, NKp44, and NKG2D	Balsamo et al., 2013
Hypoxia (short and long term) in combination with IL-15 + IL-18 or IL-2 priming	Inhibition of oxidative metabolism, induction of CXCL8, VEGF, and MIF, release of IFNγ, TNFα, GM-CSF, and little CCL3 and CCL5	Parodi et al., 2018
Hypoxia (short term) in combination with IL-15 priming	No change in glycolytic flux or glucose consumption, enhanced release of CCL3, CCL4, CCL5, reduction in K562 cell lysis, enhanced migration through ECM (hypoxia only), progression to late apoptosis (hypoxia only)	Velásquez et al., 2016
Chronic hypoxia (4 weeks at 1% O_2)	1,000× reduced proliferative capacity, reduced ATP and VEGF production, loss of CD56 driven by miR-210	Ang et al., 2010
HIF-1α	HIF-1α-deficient NK cells show impaired tumor cell killing, but promote tumor inhibition due to lower tumor infiltration and lack of presentation of VEGFR-1	Krzywinska et al., 2017
Anoxia	Reduction in NK cytotoxicity against MHC-negative mouse tumor cell line YAC-1	Loeffler et al., 1991
Hypoxia (1% O_2)	Reduction in NK cytotoxicity against K562 cells	Fink et al., 2003
Hypoxia	Reduction in expression of NKG2D, perforin and granzyme B, decrease in cytotoxicity against multiple myeloma cells, reduction in CD16 expression; IL-2 rescued these effects	Sarkar et al., 2013
Hypoxia (0.6%)	Reduction in NK cytotoxicity against K562 only when combined with lactate; limited but not impaired degranulation against myeloma cells and no change in ADCC due to hypoxia alone	Mahaweni et al., 2018a
Hypoxia (0.6%)	$NKG2A^+KIR^-$ NK cells degranulate more than $NKG2A^+KIR^+$ cells; no difference in degranulation for KIR^- NK cells with or without NKG2A	Mahaweni et al., 2018b

Chambers AM, Matosevic S. *Front Mol Biosci*. 2019;6:60

Effects of CD73 on NK cell function and metabolism

TABLE 2 | Effects of CD73 on NK cell function and metabolism.

Immunomodulatory factor	Effect	References
CD73		
CD73-produced adenosine	Impairment in activating receptor expression and expression of metabolism-related genes; inhibition of glycolysis and NK cytotoxicity	Chambers et al., 2018b
CD73	Induced expression of CD73 on NK cells upon co-culture with mesenchymal stem cells	Chatterjee et al., 2014
CD73-produced adenosine	Inhibition of IFN-γ and TNF-α expression; impairment in cytolysis	Raskovalova et al., 2005
CD73	Anti-4-1BB therapy did not alter recruitment of NK cells in $CD73^{-/-}$ mice	Chen et al., 2019
CD73	No change in anti-tumor effect of NK cells in CD73 KO mice	Wang et al., 2011
CD73 and A_{2A} receptor	Co-inhibition of CD73 and A_{2A} receptor enhanced intratumoral activity of NK cells	Young et al., 2016
CD73	Blocking CD73 enhanced cytotoxicity of NK cells against ovarian cancer cells *in vitro*	Häusler et al., 2014
CD73	TGF-β induced CD73 on MDSCs which in turn suppressed NK-mediated cytolysis of tumors	Li et al., 2017
CD73	Antibody-mediated blockade of CD73 enhanced anti-metastatic function of NK cells by blocking tumor growth	Stagg et al., 2012
CD73	Blockade of CD73 enhanced cytotoxicity and intra-tumoral infiltration of chimeric antigen receptor-NK cells *in vivo*	Wang et al., 2018
CD73-produced adenosine via A_{2A} receptor	Inhibition of NK cell maturation and anti-tumor activity	Young et al., 2018
CD73-produced adenosine	Inhibition of NK cell cytotoxicity	Raskovalova et al., 2006

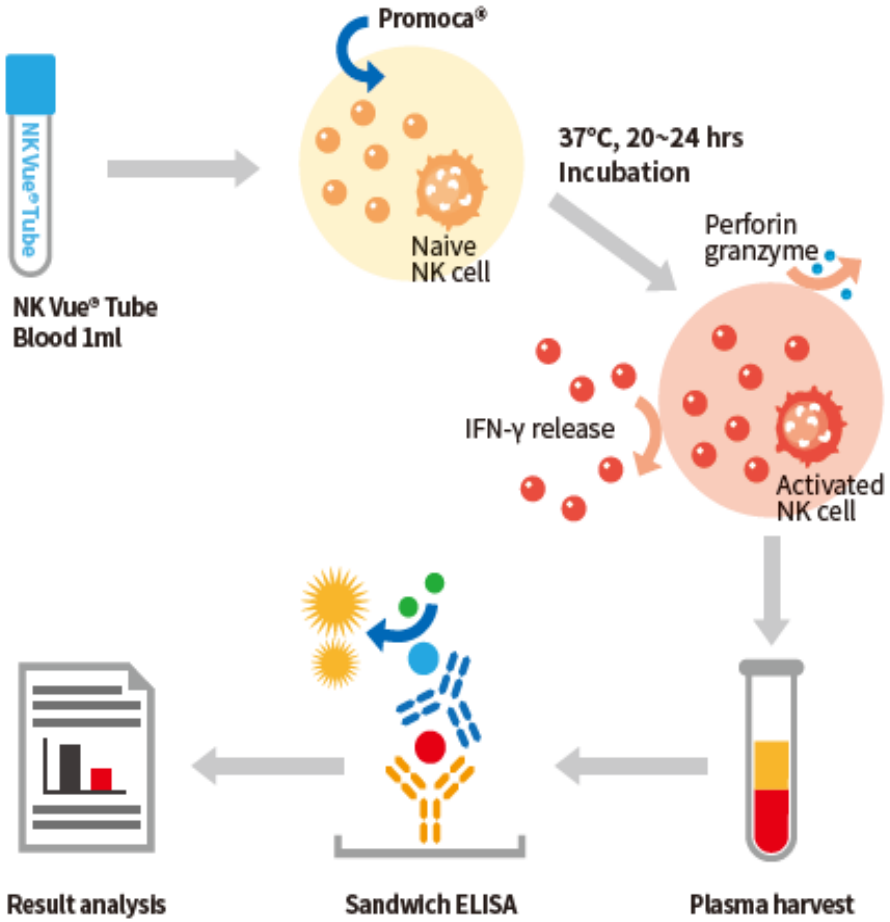
Chambers AM, Matosevic S. *Front Mol Biosci*. 2019;6:60

Methods

- A total 70 cancer patients who treated HBOT at the PARK Hospital
- Between April and October in 2020
- The concentration of interferon-γ (IFN-γ) was measured and considered to represent NKA (Natural Killer cell Activity).
- Comparison of NKA level before and after HBOT

Methods-Natural Killer cell Activity

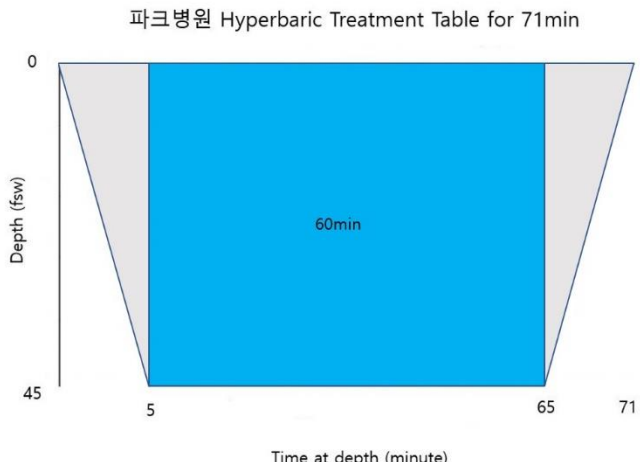
- Sampling of 1ml of whole blood
- Using special tubes for analysis with NK Vue® Gold kit
- Measuring the amount of IFN-γ using sandwich ELISA

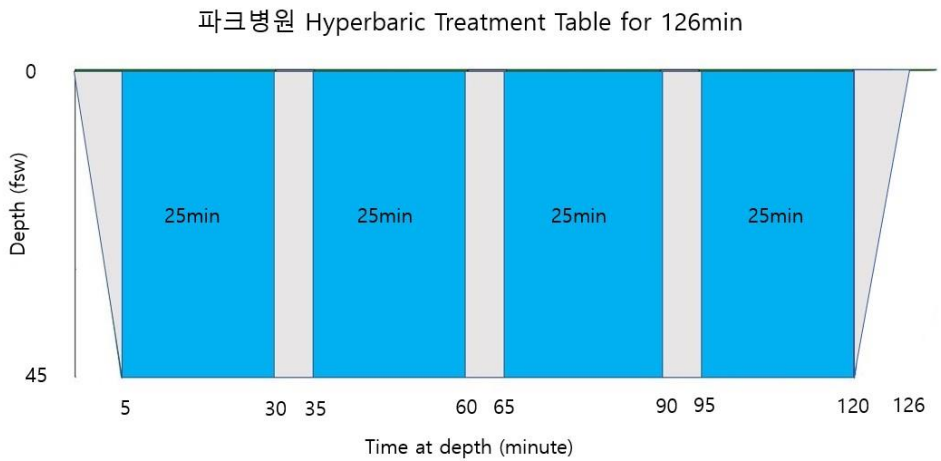


Methods-HBOT protocol

- Over 10 hyperbaric sessions
- 3~6 days a week
- 60~100 minutes each session
- 100% oxygen with air breaks at 2.4 ATA
- The hyperbaric multiplace chamber used was PVHO-1 (I-BEX, Korea).
- Oxygen was delivered by tight masks.
- Acceptable compression and decompression rates of 2.8 meter per minute were employed.

Methods-HBOT protocol





Hyperbaric chamber center

Result

Table 1. Baseline characteristics of patients

	Patients (n=70)
Age (yr)	52.4 ± 9.0
Sex (male : female)	6 : 64
Solid Cancer	
Breast cancer	32 (45.7%)
Ovary cancer	8 (11.4%)
Non-Small cell Lung cancer	7 (10.0%)
Uterine Cervical cancer	4 (5.7%)
Thyroid cancer	4 (5.7%)
Pancreas cancer	3 (4.3%)
etc.	9 (14.2%)
Hematologic malignancy	3 (4.3%)
Stage	
I	10 (14.2%)
II	12 (17.2%)
III	10 (14.2%)
IV	38 (54.2%)

Result

- Combined anti cancer drug
  - 5-Fu, Capecitabine, Paclitaxel, Cisplatin, Carboplatin, Doxorubicin, Liposomal-Doxorubicin, Gemcitabine, Irrinitecan, Ifosfamide, Leucovorin
    Albumin-bounded paclitaxel, Docetaxel, Etoposide
  - Tamoxifen, Leuprorerin, Letrozole, Fulvestrant
  - Pertuzumab, Trastuzumab, Bevacizumab, Laptinib
  - Pembrolozumab,
  - Pabociclib, Osimertinib, Crizotinib, Everolimus, Alectinib, Abemaciclib, Erlotinib

- All patients are alive.

Result

Table 2. Levels of first step NK cell activity according to the clinical characteristics

Clinical Characteristics	**NK cell activity (pg/mL)**	***P-value***
Sex		*0.026*
Male	391.17 (4 ~ 2061)	
Female	1182.70 (3 ~ 4000)	
Disease		*0.150*
Breast cancer	1363.44 (19 ~ 4000)	
Ovary cancer	1358.50 (7 ~ 4000)	
Non-Small cell Lung cancer	1071.14 (3 ~ 2061)	
Uterine Cervical cancer	110.25 (6 ~ 253)	
Thyroid cancer	1748.75 (24 ~ 4000)	
Pancreas cancer	83.00 (26 ~188)	
Stage		*0.009*
I	940.10 (19 ~ 3183)	
II	2181.25 (90 ~ 4000)	
III	440.60 (4 ~ 1807)	
IV	920.49 (3 ~ 4000)	

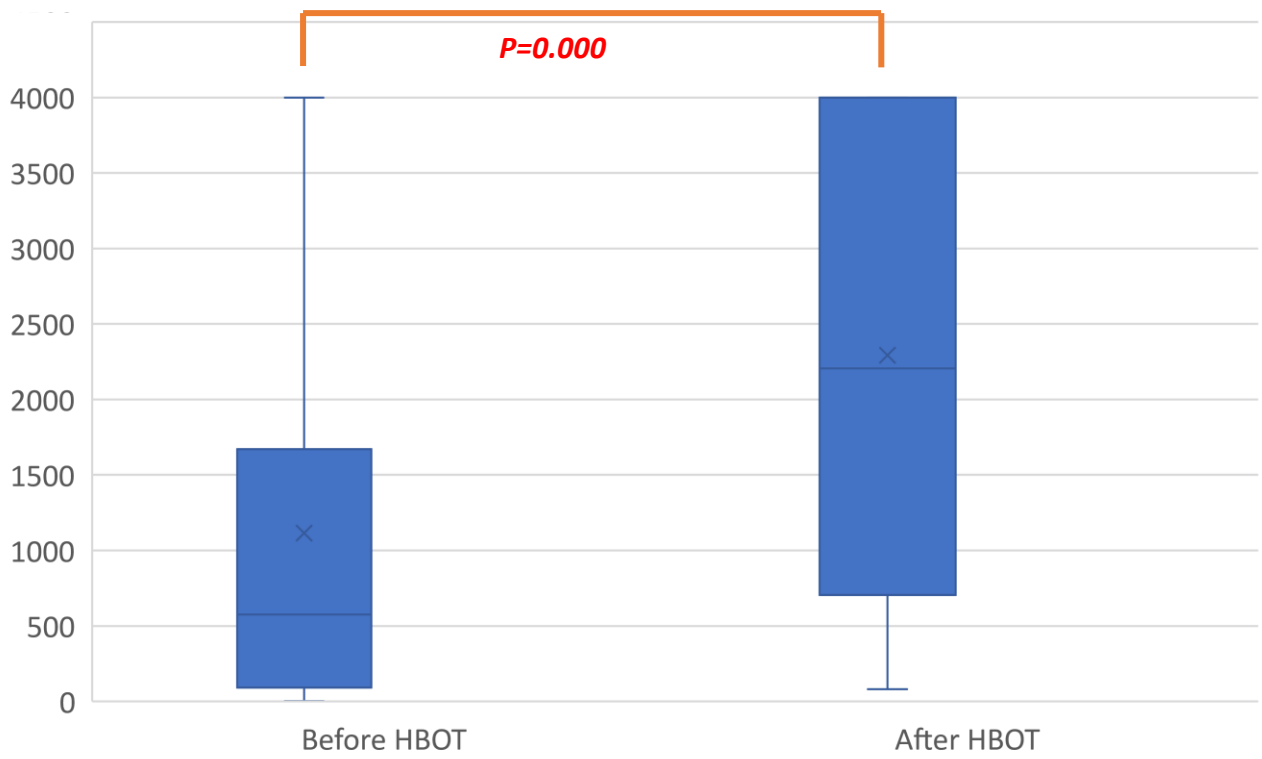
Result

Table 3. Levels of NK cell activity according to the Hyperbaric oxygen (HBO) treatment

	NK cell activity (pg/mL)	***P-value***
HBO		*0.000*
Before	1114.86 (3 ~ 4000)	
After	2293.64 (82 ~ 4000)	

Result

Figure 1. Comparison of natural killer (NK) cell activity, according to the Hyperbaric oxygen (HBO) treatment



Result

Table 4. Levels of NK cell activity according to HBOT in Breast cancer patients. (n=32, 45.7%)

	NK cell activity (pg/mL)	***P-value***
HBO		*0.000*
Before	1363.44 (19 ~ 4000)	
After	2472.78 (151 ~ 4000)	

Table 5. Levels of NK cell activity according to HBOT in Stage IV patients. (n=37, 54.2%)

	NK cell activity (pg/mL)	***P-value***
HBO		*0.000*
Before	920.49 (3 ~ 4000)	
After	2382.43 (82 ~ 4000)	

Conclusion

- HBOT stimulates immunity by promoting NKA in cancer patients.
- Further studies need for the application of HBOT for the stimulation of immunity in cancer patients.

In future

: *Aspect of oncology and immunologic stimulation*

- OS vs DFS
- Stage?
- Cancer type?
- How many session?
- Other marker? (ex. Neutrophil/Lymphocyte ratio)
- Other Oncology Pt. effect

  (Nutritional support, Post OP. nerve inj…)

Thank you for attention!