Serum Malondialdehyde level as a Predictor of Failure of Weaning from Mechanical Ventilation in the NICUs

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ABSTRACT

Background: Mechanical ventilation (MV) stands as a crucial life-saving therapeutic intervention. Optimizing the time of weaning from MV to reduce its duration and avoid failure of the process represents a major challenge. Patients on mechanical ventilation frequently encounter respiratory muscle dysfunction, complicating the weaning process. Oxidative stress plays a pivotal role in mechanical ventilation-induced respiratory muscle dysfunction. Serum malondialdehyde (MDA) arises from the oxidative breakdown of polyunsaturated fatty acids and is frequently utilized as a biomarker of oxidative stress.

Objective: The main aim of this study was to assess the efficacy of serum malondialdehyde as a contributory factor in predicting failure of weaning from mechanical ventilation in neonates.

Methods: This prospective cohort study was conducted on 31 neonates recruited from the neonatal intensive care unit at Ain Shams University Hospitals between March 2023 and November 2023. All the participating neonates were mechanical ventilation-dependent for more than 72 hours. All of them were candidates for weaning according to standard weaning protocol and have successfully passed the spontaneous breathing trial (SBT). Serum MDA levels were assessed for all participants by Enzyme-linked immunosorbent assay (ELISA) pre-SBT, at 30 minutes, and 72 hours post-SBT. Based on the definition of weaning failure, which involves the need for re-intubation within 72 hours after a successful SBT, the enrolled neonates were further categorized into two groups: those with weaning success and those with weaning failure.

Results: The weaning success group included 14 neonates, whereas the weaning failure group included 17 neonates. Serum malondialdehyde levels were significantly higher pre-SBT, at 30 minutes post-SBT, and 72 hours post-SBT in the weaning failure group compared to the weaning success group. A value exceeding 223 ng/ml was identified as a cut-off for serum MDA to predict failure of neonatal weaning from MV with a sensitivity of 94% and specificity of 100%.

Conclusion: Evaluating serum MDA levels could serve as a valuable biomarker in predicting failure of neonatal weaning from mechanical ventilation.

Keywords: Weaning, Mechanical Ventilation**,** MDA**.**

Introduction:

The term "weaning" refers to the gradual reduction of ventilator support. Spontaneous breathing trial (SBT) evaluates the patient's ability to breathe with minimal or no ventilator assistance. Initiating SBT and weaning should commence with an assessment to determine whether the underlying cause of respiratory failure has been addressed or remains unresolved ***(Zein et al., 2016)***. Optimizing the time of weaning from mechanical ventilation (MV) remains a persistent challenge. Traditionally, weaning relies on clinical judgment and considers various factors including ventilator settings and blood gas readings ***(Goel et al., 2018)***. Patients undergoing mechanical ventilation frequently develop respiratory muscle dysfunction, complicating the weaning process. There is no simple method to predict or diagnose respiratory muscle dysfunction as diagnosis relies on measurements in diaphragmatic muscle fibers ***(Levine et al., 2008)***.

Oxidative stress significantly contributes to mechanical ventilation-induced respiratory muscle dysfunction; prolonging MV time and making the weaning process difficult ***(Kevin ,2006)***. Serum malondialdehyde (MDA) is generated through the oxidative decomposition of polyunsaturated fatty acids, promoting its use as an oxidative stress biomarker ***(Grune et al., 2007)***.

Aim of the work:

This study sought to investigate the value of serum malondialdehyde as a predictor of failure of weaning from mechanical ventilation in neonates.

Ethical Consideration:

- The study was conducted after obtaining approval from the local ethics committee, Faculty of Medicine, Ain-shams University **(FMASU MS 138/2023)**
- Informed consent was obtained from caregivers of newborns before participation in the study
- All data and results are kept confidential.
- Caregivers of the participants have the right to refuse or withdraw from the study at any time.
- The authors declare that they have no conflict of interest regarding the study or the publication.
- The study and the publication are self-funded.

Sample size calculation:

The sample size was calculated according to ***Cleber Verona et al. (2015)*** who reported a large effect size (>2) comparing mean MDA level between success and failure weaning cases. Assuming a proportion of weaning success of 60%, a sample size of 31 cases with expected successful weaning in 19 achieves a power of 80% to detect a large effect size of 1.1 using a two-independent samples t-test with a level of significance of 0.05

Inclusion Criteria:

I-Full-term (gestational age > 37 weeks) and pre-term neonates (gestational age from 33-37 weeks)

II-Birth weight more than 1000 gm

III-The selected neonates required mechanical ventilation for over 72 hours within their first 28 days of life.

IV-The selected participants were ready to be weaned according to standard protocol

Exclusion Criteria:

I- Extremely low birth weight (ELBW) <1000 gm.

II-Prematurity with gestational age less than 33 weeks

III- Major upper or lower airway anomalies

IV-Congenital myopathies.

V-Severe neurological problems.

VI-Unstable patients with respiratory or circulatory failure.

Study Procedure:

This prospective cohort study was conducted on 31 full-term and pre-term neonates. They were admitted to the neonatal intensive care unit at Ain Shams University Hospitals for 9 months from March 2023 to November 2023.All participating neonates were subjected to the following:

I) Full history taking with determination of gestational age based on the date of the last menstrual period.

II) Clinical examination and hemodynamic assessment. Gestational age was confirmed using the modified Ballard score. APGAR scores at 1 and 5 minutes were recorded.

III) weaning according to the following standard weaning protocol ***(Shalish et al., 2021)***

a-. Patients on minimal ventilatory settings (SIMV; PIP ≤16 cm H2O, PEEP ≤6 cm H2O, Rate ≤20, FiO2 ≤0.30. / AC\PSV; birth weight (BW)>1000 gm: mean air way pressure (MAP) ≤8 cm H2O and FiO2 ≤0.30 / VTV: (TV) ≤4-4.5 mL/kg (5-6mL/kg if >2 weeks of age) and FiO2 ≤0.30. (HFOV): BW >1000 gm: MAP ≤9 cm H2O and FiO2 ≤0.30).

b. normal venous blood gases.

c. lung disease is stable or resolving.

d. Patients are hemodynamically stable (with little or no inotropic support).

e. Patients can initiate spontaneous breaths assessed by a SBT ***(Teixeira et al., 2021).***

IV) Laboratory Analysis:

Routine laboratory investigations were assayed before weaning and included a complete blood count (CBC) that was analyzed on Sysmex-XN-1000 (Sysmex Europe GmbH, Bornbarch, Germany), C-reactive protein (CRP) which was performed using Roche/Hitachi Cobas C501 System (Roche Diagnostics International Ltd., Switzerland), venous blood gas which was done using GEM premier 3500 (Instrumental Laboratory Company-Bedford, USA). Serum MDA levels were measured at three time points: (i) pre SBT (ii) 30 minutes post SBT and (iii) 72 hours post SBT. Serum MDA levels were assayed using a commercially available double—antibody sandwich (non-competitive) enzyme-linked immunosorbent assay (SinoGeneClon Biotech Co.,Ltd, Shanghai, China, Catalog no: SG-00097) according to the manufacturer's protocol**.**

V) Lastly, the enrolled neonates were categorized into two groups after a successful SBT:

A) weaning success neonates: neonates who didn't need further reintubation within 72 hours of successful SBT

B) weaning failure neonates: neonates who needed reintubation and resumption of ventilatory support within 72 hours of successful SBT

Statistical Analysis

Data were collected, coded, and analyzed with a statistical package for social sciences, (IBM SPSS) version 23. Quantitative data were presented as a mean± standard deviation (SD) in parametric distribution and median with inter-quartile range (IQR) in non-parametric distribution. Qualitative data were presented as frequency and percentage and analyzed by the Chi-square test. The comparison between two groups regarding quantitative parameters was done by using independent t-test for data with parametric distribution, and Mann-Whitney test for data with non-parametric distribution. Spearman correlation coefficients were used to assess the correlation between two quantitative parameters in the same group. The p-value was considered non-significant if more than 0.05, significant if less than 0.05, and highly significant if less than 0.01. The receiver operating characteristic (ROC) curve was used to determine the best cut-off point, sensitivity, and specificity.

Results

Table (1): Clinical characteristics of the studied groups

		Weaning Success (Number: 14)	Weaning Failure (Number: 17)	Test value	P-value
APGAR 1 minute	Median (IQR)	6 (6 – 7)	5 (5 – 6)	-2.390≠	0.017*
APGAR 5 minutes	Median (IQR)	8 (8 – 9)	8 (7 – 8)	2.149≠	0.032*
Gestational age (weeks)	Mean ± SD	37.10 ± 1.44	35.42 ± 1.58	3.024•	0.005**
Gender Number (%)	Female Male	6 (42.9%) 8 (57.1%)	4 (23.5%) 13 (76.5%)	1.312¶	0.252
Birth Weight (kg)	Mean ± SD	2.57 ± 0.65	2.53 ± 0.72	0.170•	0.866
Postnatal age at Intubation (days)	Median (IQR)	3 (2 – 6)	7 (3 – 11)	-1.329≠	0.184
Postnatal age at Extubation (days)	Median (IQR)	12 (8 – 24)	10 (7 – 17)	-0.239≠	0.811
Mortality rate Number (%)	Survived Died	14 (100.0%) 0 (0.0%)	10 (58.8%) 7 (41.2%)	7.446¶	0.006**

¶: Chi-square test; •: Independent t-test; ≠: Mann-Whitney test

*: Significant, **:Highly significant

The enrolled neonates were categorized into two groups according to the need for re-intubation within 72 hours of a successful weaning trial. The first group comprised 14 neonates who were successfully weaned from mechanical ventilation. The second group comprised 17 neonates who

experienced failure of weaning from MV. The clinical characteristics of both groups are presented in **table 1.** It revealed that weaning failure neonates have significantly lower Apgar scores at 1 and 5 minutes and gestational ages than weaning success neonates. Moreover, the mortality rate was significantly higher in the weaning failure group.

Table (2): Ventilation data of the studied groups

Ventilation Data		**Weaning Success (Number: 14)**	**Weaning Failure (Number:17)**	**Test value**	**P-value**
Indications of MV Number (%)	Sepsis (multiple organ failure) RDS	4 (28.6%) 10 (71.4%)	8 (47.1%) 9 (52.9%)	2.287¶	0.319
Duration of MV (Days)	Median (IQR)	5 (2 – 7)	7 (5 – 14)	-2.218≠	0.027*
Mode of MV Number (%)	HFOV SIMV	2 (14.3%) 12 (85.7%)	8 (47.1%) 9 (52.9%)	3.774¶	0.52

HFOV*: high-frequency oscillatory ventilation;* **SIMV***: synchronized intermittent mandatory ventilation.*

¶: Chi-square test; ≠: Mann-Whitney test

*: Significant

Table (2) presents the ventilation data of the studied groups. It shows that neonates experiencing weaning failure had significantly longer periods of mechanical ventilation as compared to those who were successfully weaned.

Table (3): Comparison between weaning success and failure groups regarding routine laboratory parameters

		Weaning Success (Number: 14)	**Weaning Failure (Number:17)**	**Test value**	**P-value**
HGB (g/dl)	Mean ± SD	13.57 ± 2.15	11.62 ± 1.73	2.798•	0.009**
TLC ($\times 10^3/\mu L$)	Median (IQR)	9.5 (8 – 13)	17 (8 – 22)	-1.590≠	0.112
PLTS ($\times 10^3/\mu L$)	Median (IQR)	259 (215 – 300)	219 (109 – 290)	-1.290≠	0.197
CRP (mg/L)	Median (IQR)	33.5 (3 – 50)	31 (9 – 70)	-0.715≠	0.474
pH	Mean ± SD	7.32 ± 0.04	7.21 ± 0.09	4.088•	0.000**
pCO2 (mmHg)	Mean ± SD	47.02 ± 7.85	55.94 ± 8.01	3.111•	0.004**
HCO3 (mmol/L)	Mean ± SD	19.05 ± 3.70	20.53 ± 5.09	-0.907•	0.372

HGB*: hemoglobin;* **TLC***; total leucocytic count;* **PLTS***: platelets* **CRP***: c-reactive protein*

•: Independent t-test; ≠: Mann-Whitney test

**: Highly significant

Table (3) compares weaning success and failure groups regarding routine laboratory parameters. There is a significant decrease in hemoglobin levels in weaning failure as compared to weaning success neonates. Moreover, the weaning failure group showed a significant reduction in PH and an increase in PCO2 as compared the weaning success group.

Table (4): Comparison between weaning success and failure groups regarding serum MDA pre-SBT, 30 minutes post and 72 hours post-SBT

Serum MDA		**Weaning Success**	**Weaning Failure**	**Test value**	**P-value**
		Number = 14	**Number = 17**		
pre SBT (ng/ml)	Median (IQR)	163.05 (146.6 – 199.2)	325.9 (293.9 – 353.3)	-4.684≠	0.000**
30 mins post SBT (ng/ml)	Median (IQR)	124.2 (78.76 – 141.3)	627 (581 – 886)	-4.725≠	0.000**
72 hours post SBT (ng/ml)	Median (IQR)	63.96 (9 – 99.78)	2000 (1957 – 2000)	-4.782≠	0.000**

≠: Mann-Whitney test

**: Highly significant

The comparison between weaning success and failure groups regarding serum MDA levels pre-SBT, 30 minutes post-SBT, and 72 hours post-SBT is illustrated in **table (4).** It shows that serum MDA levels at the three points were significantly higher in weaning failure neonates than weaning success ones.

Table (5): Correlation between pre-SBT serum MDA levels and other studied parameters

	S.MDA (pre SBT)	
	r§	**p-value**
Gestational age (in weeks)	-0.047	0.801
Age of admission	-0.052	0.781
Age of Intubation	0.149	0.422
Age of Extubation	0.140	0.452
Weight (in kg)	0.073	0.698
O2 saturation	-0.756	0.000**
Days of MV	0.358	0.048*
HGB	-0.507	0.004**
TLC	0.289	0.115
PLTS	-0.317	0.082
CRP	0.280	0.127
pH (pre-extubation)	-0.715	0.000**
pCO2 (pre-extubation)	0.559	0.001**
HCO3 (pre-extubation)	0.208	0.262

§Spearman correlation coefficients

*: Significant, **: Highly significant

Figure (1): Receiver operating characteristic (ROC) curve for pre-SBT serum MDA to detect failure of weaning.

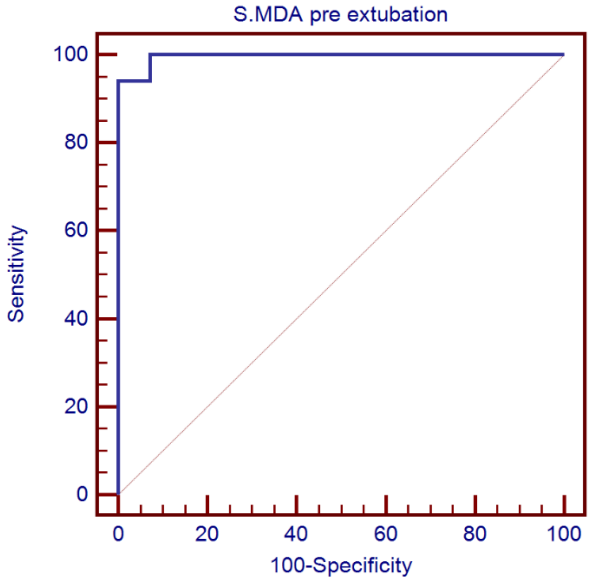

Table (6): Cut-off point, Sensitivity, Specificity, PPV, and NPV of pre-SBT serum MDA to predict failure of neonatal weaning from mechanical ventilation.

Cut off point	AUC	Sensitivity	Specificity	PPV	NPV
>223	0.996	94.1	100.0	100.0	93.3

AUC: Area under curve; **PPV:** Positive predictive value; **NPV:** Negative predictive value

The receiver operating characteristic (ROC) curve for pre-SBT serum MDA levels showed that the best cut-off point to predict failure of weaning was > 223 ng/ml with a sensitivity of 94.1%, specificity of 100.0%, and area under the curve (AUC) of 0.996; as shown in **figure 1 and table 6.**

DISCUSSION

Weaning from mechanical ventilation entails the comprehensive process of liberating the patient from mechanical assistance and discontinuing the endotracheal tube. Managing these patients poses a major challenge, as the pathophysiologic mechanisms responsible for weaning failure are still poorly understood ***(Tobin et al.,2006)***. This study was conducted to investigate the value of serum MDA as a predictive marker of the outcome of neonatal weaning from invasive mechanical ventilation.

Our data confirmed lower gestational age in the weaning failure group, which copes with several previous studies which reported that a

decreased gestational age raises the likelihood of weaning failure. Respiratory muscle weakness, airway abnormalities, patent ductus arteriosus, and inadequate respiratory control are believed to contribute to weaning failure in infants with lower gestational age ***(Manley et al.,2016 and Kidman et al.,2021).***

In the present study, the group that accomplished successful weaning demonstrated higher APGAR scores at 1 and 5 minutes. This finding aligns with the research of ***Hermeto et al., (2009)*** which reported that successfully extubated infants exhibited higher Apgar scores compared to those who experienced weaning failure**.**

In addition, our study reported that prolonged intubation was associated with a higher risk of weaning failure. These results are consistent with ***Bruhn et al., (2001)*** and ***Shin et al. (2017)*** studies which found that weaning failure was linked to extended duration of mechanical ventilation and ICU length of stay**.**

Our results proved that the weaning failure group had a statistically significant decrease in PH and an increase in pCO2 levels as compared to the weaning success group. Our findings coped with previous studies done by ***El-Beheidy et al. (2018)*** and ***Keyal et al. (2018)*** who concluded that ABG can be a valuable tool in assessing patients' response to weaning from MV. Studies indicate that the main contributors to neuromuscular fatigue primarily stem from muscle fibers and are mainly linked to energy depletion or pH variations caused by lactate accumulation ***(Westerblad et al. 1991 and Vassilakopoulos et al. 1998)***

Oxygen-derived free radicals have been suggested to play a role as mediators of diaphragm dysfunction ***(Anzueto et al.,1994)***. Previous studies conducted on the diaphragms of critically ill patients receiving MV have identified elevations in oxidative stress, muscle fiber atrophy, and injury ***(Funk et al.,2010)***.Upon evaluating the oxidative stress marker "MDA"; our study demonstrated a statistically significant increase in its serum levels at 3 points: pre-, at 30 minutes, and the end of 72 hours post-SBT in the weaning failure group as compared to the weaning success group. This agreed with ***Verona et al. (2015)*** study which proved lower pre-SBT levels of nitric oxide and higher pre-SBT levels of both MDA and vitamin C in weaning failure patients.

In our current study, the best cut-off value for pre-SBT serum MDA levels to predict weaning failure was found to be >223 ng /ml with a specificity of 100% and sensitivity of 94.1%. Serum MDA levels showed a significant positive correlation with both the duration of MV and pre-SBT pCO2, in addition to a significant negative correlation with the following parameters: oxygen saturation, hemoglobin, and pH. From the previous data, the suggested valuable role of serum MDA as a

predictor of the outcome of weaning from MV has emerged.

According to previous studies on incidence and outcome of weaning from mechanical ventilation; hospital mortality was found to be higher for patients with prolonged weaning compared with patients with simple weaning .Also, they found that any unnecessary delay in extubating patients who have successfully passed a weaning test may raise mortality due to complications associated with extended mechanical ventilation ***(Lorenate et al. 2015 and Sangsari et al., 2022)***.This explains why neonatologists seek to wean neonates from mechanical ventilation as soon as possible and hence highlighting the beneficial role of a sensitive and specific marker in predicting the outcome of weaning from mechanical ventilation.

CONCLUSION:

Assessing serum MDA levels can serve as a valuable marker, offering high specificity and sensitivity in predicting the outcome of neonatal weaning from mechanical ventilation.

LIMITATIONS:

Our study has some limitations as small sample size and lack of serial measurements of serum MDA for extended durations after neonatal weaning.

RECOMMENDATIONS:

Additional larger sample size studies are necessary to either substantiate or refute our findings. The inclusion of serial measurements of MDA levels for extended durations to elucidate its significance in the follow-up of extubated neonates and to be correlated with different morbidities and with patient mortality

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REFERENCES

Anzueto A, Supinski G, Levine S, et al. (1994): Mechanisms of disease: are oxygen-derived free radicals involved in diaphragmatic dysfunction? Am J Respir Crit Care Med;149: 1048–52.

Bruhn A, Apablaza F, Bernucci F, Segovia V, Zúñiga P, Hernández G, et al. (2001): Impact of weaning failure in the evolution of patients under mechanical ventilation. Critical Care; 5(1):21.

El-Beheidy RM, Abdulrahman DA, Bayoumi SS, Hassan WA. (2018): Prediction of weaning failure of mechanical ventilation using clinical parameters in PICU. Zagazig University Medical Journal ;24(5):386-93.

Funk GC, Anders S, Breyer MK, Burghuber OC, Edelmann G, Heindl W, et al. (2010): Incidence and outcome of weaning from mechanical ventilation

according to new categories. European Respiratory Journal;35(1):88-94.

Goel N, Chakraborty M, Watkins W, Banerjee S. (2018): Predicting Extubation outcome – A model incorporating heart rate characteristic index. The Journal of Pediatrics; 195:53-58.

Grune T, Berger MM (2007): Markers of oxidative stress in ICU clinical settings: present and future. Curr Opin Clin Nutr Metab Care, (6)712-7.

Hermeto F, Bottino MN, Vaillancourt K, Sant'Anna GM. (2009): Implementation of a respiratory therapist-driven protocol for neonatal ventilation: impact on the premature population. Pediatrics; 123(5):907-916.

Kevin L. (2006): Comment on "Effects of short vs. prolonged mechanical ventilation on antioxidant systems in piglet diaphragm". Intensive Care Medicine; 32:1446.

Keyal N, Shrestha G, Amatya R, Shrestha P, Acharya S, Marhatta M, et al. (2018): 1117: Influence of Abg To Guide Extubation In Icu Patients After Spontaneous Breathing Trial. Critical Care Medicine; 46(1):542.

Kidman AM, Manley BJ, Boland RA, Davis PG, Bhatia R. (2021): Predictors and outcomes of extubation failure in extremely preterm infants. Journal of paediatrics and child health. ;57(6):913-9.

Levine S, Nguyen T, Taylor N, Friscia ME, Budak MT, Rothenberg P, Zhu J, Sachdeva R, Sonnad S, Kaiser LR, Rubinstein NA. (2008): Rapid disuse atrophy of diaphragm fibers in mechanically ventilated humans. New England Journal of Medicine. Mar 27;358(13):1327-35.

Lorente L, Martín MM, Abreu-González P, Ramos L, Argueso M, Cáceres JJ, et al. (2015): Association between serum malondialdehyde levels and mortality in patients with severe brain trauma injury. Journal of Neurotraum; 32(1):1-6.

Macintyre NR. (2012): Evidence-Based Assessments in the ventilator Discontinuation process. Respiratory Care; 57(10):1611–1618.

Manley BJ, Doyle LW, Owen LS, Davis PG. (**2016):** Extubating extremely preterm infants: predictors of success and outcomes following failure. J Pediatr.;173:45–9.

Sangsari R, Saeedi M, Maddah M, Mirnia K, Goldsmith J. (2022): Weaning and extubation from neonatal mechanical ventilation: an evidenced-based review. BMC Pulmonary Medicin; 22: 421.

Shalish W, Sant's Anna G, Keszler M. (2021): Weaning and extubation from mechanical ventilation.Assisted ventilation of the Neonate; 26:304**.**

Shin H, Chang J, Ahn S, Kim T, Park C, Lim J. (2017): Clinical factors associated with weaning failure in patients requiring prolonged mechanical ventilation. Journal of thoracic disease; 9(1):145-151.

Teixeira R, Carvalho A, Araujo R, Veloso F, Kassar S, Medeiros A. (2021): Spontaneous Breathing Trials in Preterm Infants; Systematic Review and Meta-Analysis. Respiratory care; 66(1):129–137.

Tobin MJ, Jubran A. (2006): Weaning from mechanical ventilation. In: Tobin MJ, editor. Principles& practice of mechanical ventilation.2nd ed. New York: McGraw-Hill; pp.1185–220.

Vassilakopoulos T, Zakynthinos S, Roussos C. (1998): Muscle function. In: Marini JJ,Slutsky AS, editors. Physiological basis of ventilator support. New York: Marcel Dekker; pp. 102–52.

Verona C, Hackenhaar F, Teixeira C, Medeiros T, Alabarse P, Salomon T et al. (2015): Blood markers of oxidative stress predict weaning failure from mechanical ventilation. Journal of Cellular and Molecular Medicine**;** 19(6):1253-1261.

Westerblad H, Allen D. (1991): Changes of myoplasmic calcium concentration during fatigue in single mouse muscle fibers. J Gen Physiol ; 98: 615–35

Zein H, Baratloo A, Negida A, Safari S. (2016): Ventilator Weaning and Spontaneous Breathing Trials; an Educational Review. Emergency; 4(2): 65–71.