

Conference Paper

Postmortem Interval Estimation Time from Algor mortis Temperature of Rats Expressed by MARS Model Approach

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Abstract

Estimation of Postmortem is one of the challenges in forensic science. The aim of this study was to construct a MARS model of Postmortem interval estimation time (PMT) from algor mortis temperature in Rat. Sixteen healthy male rats (*Rattus norvegicus*), onemonth old and weigh 100 gram were randomly divided into two groups (eight/each group) and were acclimated respectively among the ambient room (temperature over 28°C) and at the conditioning room (temperature over 20°C). The animals then were sacrificed in two days (four rats/day for each divided room) then algor mortis by rectal temperature were recorded after death at 0 and 2,4,6,8, 10,12, 14,16, 18,20 till 22 h respectively. The MARS model is nonlinear regression but performed as a multilinear curve that can have splines fitting and be defined as function model $Y = 35.321 + 1.253 * BF_1 + 0.436 * BF_2 - 1.319 * BF_3$; and on 20°C condition room as $Y = 29.980 + 1.354 * BF_1 + 0.799 * BF_2 - 1.347 * BF_3$. Therefore, performance model was comprised by multilinear curve, then function model of algor mortis on ambient room be defined into three PMT intervals i.e: 1) $Y = 37.94 - 0.11 * (0-2h)$ ($p > 0.00$); 2) $Y = 40.88 - 1.87 * (2-6h)$ ($p < 0.00$) and 3) $Y = 30.82 - 0.09 * (6-22h)$ ($p < 0.00$) while on 20°C condition room, was: 1) $Y = 34.78 - 0.09 * (0-2h)$ ($p < 0.00$); 2) $Y = 37.97 - 2.38 * (2-6h)$ ($p < 0.00$) and 3) $Y = 25.36 - 0.04 * (6-22 h)$ ($p > 0.00$). The acceleration of the declining algor mortis at conditioning room showed steeper than on ambient room at 2-6h PMT interval (β : 2,38 vs 1,87). Postmortem Time Interval Estimation from Algor mortis Temperature of Rats could be expressed by MARS Model. The pattern model of estimation comprised by multilinear curve with splines was fitted at both of the experimental rooms.

Keywords: Postmortem time interval, algor mortis, MARS model estimation.

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1. Introduction

Some forensic Researches have been attempted in order to evaluate time since death (TSD). The correlation of postmortem time (PMT) interval with body cooling after death (algor mortis) have been established and confirmed in forensic studies. In most cases, soon after death occurs, the body temperature began to drop and continued until the internal temperature reaches the temperature of the environment. Rodrigo [1] use the Laplace transform to construct a method for estimating TSD from algor mortis temperature. The author gave a recent overview of different concept and techniques which includes Rainy (the author of nineteenth century), Knight, Nokes and Brown method, Henssge *et al.*, Green *et al.*, method and then the well-known Marshall-Hoare's model. The author of the nineteenth century made the first attempt to introduce the mathematical concepts. They used sequential rectal temperatures of corpses to calculate the rate of postmortem body cooling per hour and stated that it was not absolutely correct when applied to human bodies. However, Marshall's research clearly established that postmortem body cooling is not a single exponential expression, but instead, it is a curve that has a sigmoid configuration with an initial temperatur lag [1, 2].

The decrease of postmortem temperature is mainly determined by physical condition. Dying and death are consecutive final biological process. After death occurs, tissue metabolism does not cease immediately, but will still continue instead for some hours [3]. The function of three systems i.e. the circulatory, respiratory and nervous system are crucial to the life of the organism. After the irreversible cessation of respiratory or circulatory arrest of the blood flow through the whole body is diminished and the tissue will fail to receive the oxygen. And biological death will occur, which the cell of organism begin to die, is distinguished as well. The next stage is individual death, when brain and brainstem death takes place [4]. With irreversible circulatory arrest, all muscles will become completely flaccid due to loss of tone [3], accompanied with the cessation of hypothalamic which failed to perform the thermoregulation function. Much body heat are generate through the skeletal muscles and these structures are the primary importance for increasingly metabolic rate and maintaining constant body temperature [5]. Postmortem interval estimation is very important task in forensic study. Development of concepts and techniques in using postmortem body cooling temperature is often used in estimation of the TSD. This assumption is not valid as can be verified from experimental result [1, 6]. The aim to this study was to construct a mathematical Multivariate Adaptive Regression Splines (MARS) model [7, 8] of PMT

interval estimation from algor mortis (postmortem body cooling temperature) in males Rat, depend on the room at ambient temperature and condition temperature.

2. Materials and Methods

This study was undertaken from September to Desember 2016. Sixteen clinically healthy male rats (*Rattus norvegicus*), one month old and weigh 100 gram were assigned to be the animal model. The rats were randomly divided into two groups (eight/each group) and were acclimated respectively among the ambient room (temperature over 28°C) and at the conditioning room (temperature over 20°C) for one week before starting the experiment. The animals then were sacrificed with anesthesia using ketamin, in two days (four rats/day for each divided room between 0 to 22 hours after death). Then, algor mortis by rectal body temperature were recorded from each rat cadaver immediately at 0 and 2,4,6,8,10,12,14,16,18,20 till 22 h postmortem respectively. The temperature measurement were taken by digital thermometer.

The collected data were analyzed by using MARS. The MARS model aims to identify the relationship between the PMT and algor mortis pattern on the ambient room or conditioning room is nonlinear regression but performed as a multilinear curve that can have splines fitting. The Result of MARS analysis PMT estimation model are: $Y = B_0 + B_1 * BF_1 + B_2 * BF_2 + \dots + B_k * BF_k$ where Y: Algor mortis (postmortem body cooling temperature); B_0 : temperature constant; $B_1 \dots B_k$: spline Coefficient Regression; $BF_1 \dots BF_k$: Basic function no 1,2.....no. k (=time interval). Criteria of the best fitting MARS estimation Model assigned by minimizing the GCV (Generalized cross-validation criterion) [7]. If the model's performance was comprised by multilinear curve with spline, the data was analyzed by *Regression Product Moment*. T-test was used to determine the validity of algor mortis pattern in the time interval of death among those both difference room. Additionally, if the finding model showed that there is a significant difference ($t > t_{table}$, with alfa 0.05), the next steps will be analyzing the knot of algor mortis in both of those room by comparing the regression coefficient (R^2) and calculating the difference using t-test [8]. The data was calculated using Multivariate Adaptive Regression Splines (MARS) and were performed by MARS 2.0 dan XL stat 14 of statistical computer software.

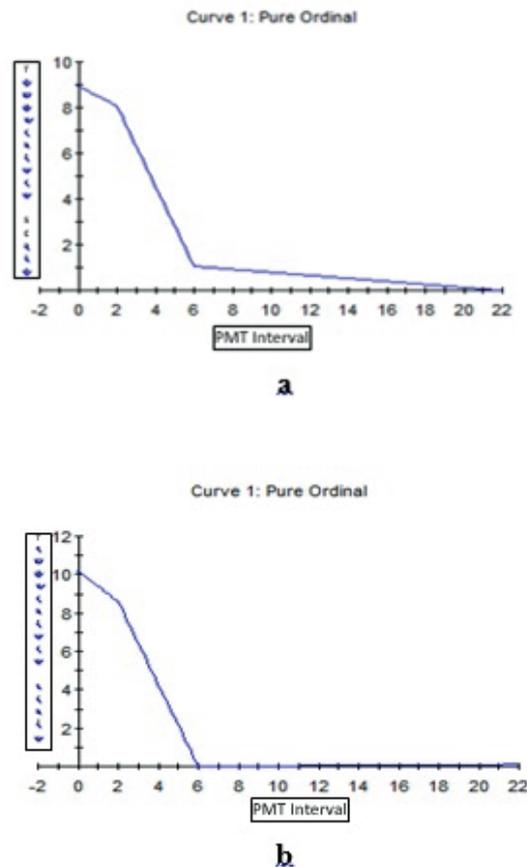


Figure 1: The Relation of Postmortem Time (PMT) Interval with Algor mortis Temperature of Rat (PMT interval were 0-2 h; 2-6 h and 6-22h) a. MARS Model for Algor mortis on ambient temperature room over 28°C b. MARS Model for Algor mortis in 20°C condition room.

3. Result and Discussion

This result of MARS analysis, performed a model of estimation of the PMT interval from algor mortis temperature (among the ambient room and conditioning room) comprised by multilinear curve with splines as follow in Figure 1. Algor mortis temperature of rat was detected declining in the same pattern ($p < 0,001$) not only at the ambient room but also at the conditioning room.

MARS Model for Algor mortis on ambient room, was obtained with these terms and conditions i.e. $MO = 0$, $MI = 1$, $BF = 4$, with Y function = $35.321 + 1.253 * BF_1 + 0.436 * BF_2 - 1.319 * BF_3$; where $BF_1 = \max(0, \text{hour} - 6.000)$; $BF_2 = \max(0, 6.000 - \text{hour})$; $BF_3 = \max(0, \text{hour} - 2.000)$; $GCV = 0.273$ (curve 1a) MARS Model for Algor mortis in conditioning room, was obtained with these terms and conditions: $MO = 0$, $MI = 1$, $BF = 4$, With Y function = $29.980 + 1.354 * BF_1 + 0.799 * BF_2 - 1.347 * BF_3$; where $BF_1 = \max(0, \text{hours} - 6.000)$; $BF_2 = \max(0, 6.000 - \text{hours})$; $BF_3 = \max(0, \text{hours} - 2.000)$; $GCV = 2.202$ (curve 1b)

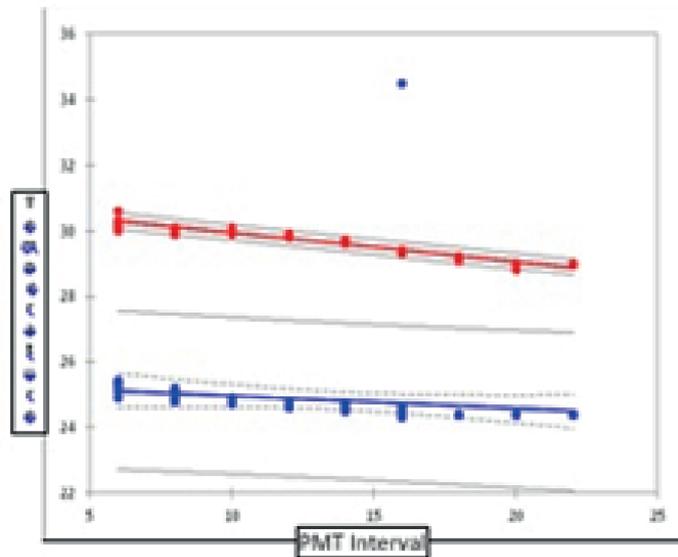


Figure 2

TABLE 1: MARS Model Postmortem Time (PMT) Interval Estimation with Algor mortis Temperature of Rat on Ambient Room and Conditioning Room.

PMT Interval	MARS Estimation Model	
	Ambient Room	Conditioning Room
0–2 h	$Y = 37,94 - 0.11 \text{ *hour*}$	$Y = 34.78 - 0.09 \text{ *hour**}$
2–6 h	$Y = 40.88 - 1.87 \text{ *hour**}$	$Y = 37.97 - 2.38 \text{ *hour**}$
6–22 h	$Y = 30.82 - 0.09 \text{ *hour**}$	$Y = 25.36 - 0.04 \text{ *hour*}$
$Y = \text{algor mortis temperature}$		
Body cooling decrease not significant ($p > 0.05$)		
Body cooling decrease significant ($p < 0.0001$)		

The result of the next step analyzed that on 2-6 h PMT interval, the decline was accelerated of algor mortis at conditioning room steeper than at ambient room (2,38 vs 1,87).

The Accelerated Decline of Algor Mortis at Ambient Room (above β : 1,87) and at Conditioning Room (below: β : 2,38) and in 2-6 h PMT Interval

Furthermore, considering the MARS Estimation as multilinear curve model with the splines, there are several formulation for implementing the each spline relation piecewise, which has shown on different PMT interval, defined as in table 1.

In our study, the algor mortis pattern of rat cadaver at 0 to 2h PMT interval suggests that the rate was not significantly decreased ($p > 0,005$) at ambient room (28°C), conversely at conditioning room (20°C) were observed a significant decrease ($p < 0,001$).

The algor mortis pattern of this study, according to the Marshall and Hoare’s research, clearly established that the slow rate in the first phase and last approximately 2- 5 h.

The authors call it an initial phase or temperature plateau that is the lag period during which the body temperature remains relatively constant. The temperature detected by rectal measurement as the best representation for sigmoid shape of the cooling curve later [2, 3].

The algor mortis pattern in 0 to 2 h PMT interval, exposed the term, that dying and death are continuous final biological processes. Moreover, Madea and Kernbach-Wighton [3] suggested that the temperature plateau was mainly determined by physical preconditions as it is due to the fact that central axial temperature cannot begin to decrease until a heat gradient was set up between the core of the body with its surface. Brown and Marshall's proposition suggested that the slow algor mortis postmortem cooling rate is due to the continuing production of the heat by the internal source of the body and removed by convection from the skin. It seems to indicate that the heat produced after the death contributes, but not totally. Irreversible respiratory or circulatory arrest is the main criterion for death, followed by the early postmortem changes that predominantly body cooling [1].

The intermediate phase of our study was the investigation at 2- 6 h PMT interval, showed there was a rapid drop of the rate of cooling body, significantly decreased ($p < 0,001$) on both of the experimental rooms. Furthermore, the statistical analysis confirmed that the pattern of decreasing was steeper at conditioning room 20°C than at ambient room over 28°C (β : 2.38 vs 1.87). The result pattern of this study is according to the Marshall and Hoare's sigmoid configuration curve expression [3]. Immediately after the death, the corpses cool slowly at a uniform rate, then followed by a straight portion of varying length and slope corresponding to the period of the quickest cooling body. It suggests that before 6 h after death, the tissue metabolism still has not ceased yet, but will continue for some hours by the next stage during which the cell of organism begins to die, is considered as the cessation of brain and brainstem function [4].

With irreversible respiratory and circulatory arrest, skeletal muscles will become completely flaccid due to loss of tone. In the early postmortem, ATP can be resynthesized via creatinine kinase reaction and anaerobic glycolysis [3]. Once ATP level has fallen, actin and myosin filaments form a rigor-complex and initiated muscle fibers that have already become stiff. Postmortem heat production is low as anaerobic glycolysis ceases within a few hours postmortem. The transport process of heat release from internal source into the body surface firstly occurred convectively and consequently was continued conductively [2]. In the immediate postmortem, dissolution of rigor mortis and livor mortis is due to protein degradation. The breakdown of tissues by

autodigestive cellular enzymes were occurred in the early stage decomposition (autolysis). The phenomenon may be seen up to 6 h postmortem [3, 9].

Saber and Ali [10] noted that the forensic experts have long recognized that decomposition is initiated by a process called autolysis which induces detrimental alterations in the cell leading to cell death. The autolysis is the other criteria of the postmortem changes. The forensic investigators attention also has focused on biochemical changes of postmortem. Biochemical changes could be used as markers for times of death determination. The accurate determination including loss of selective membrane permeability, protein RNA and DNA degradation or loss of RNA transcription after death.

Moreover, Saber and Ali [10] using the analysis of the mRNA expression of FasL, Caspase-3 and Phosphatase and Tensin (PTEN) on chromosome 10 of gastrocnemius muscle of Rat. The results suggest that the three expression of genes depicted a gradual increase beginning 2 h to 4h and optimum in 6 h and then a marked decrease at 8 h after the death. Indeed, as the cancer, stem cells and genetic studies that published between 1997-1999 have found that FasL is a necrosis factor that initiate cell death; Caspase-3 is an enzyme in sharing and activate the apoptosis and PTEN plays a vital role in regulation of cell growth and apoptosis.

Thus, Saber and Ali's study (2016) of these correlation between expression level of the three genes in 2-6 h time since death, confirm the implicated to the pattern of decreasing postmortem cooling rate at the same time in our study.

In the terminal phase during 6-22 h PMT stages, the MARS model of our study described that the cooling of the body became gradually decreased, linear with increasingly postmortem time. The cooling decrease was significant ($p < 0,001$) at ambient room, conversely on conditioning room it was observed not significantly decrease ($p > 0,001$).

Rodrigo (2015) supports the recent researcher proposed exponential model which was found the plateau and a sigmoidal shape cooling curve. These forensic experts described that a slow falling curve of gradually decreasing gradient was occurred at the final stage, similar to that usually associated with cooling expressed by Newton's Law. Most of the heat removed from internal layer of the deceased body is due to convection from the skin according to a Newton's law of cooling. Earlier, in 1974 Brown and Marshall proposed, as the surface heat is lost and the outer layer of decay body start to cool, heat immediately flowing out from any subjacent layer, or from the hotter region of the body into the outer or superficial layer by conduction. The body temperature slowly dropped continuously as the core temperature approaches that of the environment and replaced the constant cooling rate and stagnant by a time [1].

The changes which occur if rigor phase broken before it reaches maximum development of flaccidity followed by onset of generalized muscle stiffening. The decrease of ATP in this periode is solely due the accumulation of intracelluler lactic acid and anaerobic breakdown of carbohydrat, fat and protein, there has been an extensive cellular damage related to biological death. [3, 10–12]. Zou and Byard (2011) reviewed forensic research's result stated that decomposition refers to process of tissue breakdown by self-digestion of the endogenous enzymes activity. Cessation of blood flow leads to the accumulation of waste products from damage cells, compromising cell and lysosomal membrane integrity, and reducing plasma and cytoplasmic pH. Moreover, the rupture of lysosomal membrane leads to release of hydrolytics enzymes and then autolytic changes progress in death body. This process usually appeared between 12-18 h in ambient temperature.

In our study, the terminal phase of algor mortis have found that the corpses cools slow at uniform rate, linear with increasingly postmortem time and then stagnant at 24.4°C in 18 h PMT interval in conditioning room and at 29°C in 22 h PMT interval for cadaver in enviromentally room. This result showed that the constant and stagnant temperature algor mortis earlier replaced on cold room.

4. Conclusions

Postmortem Time Interval Estimation from Algormortis Temperature of Rats could be expressed by MARS Model. The pattern model of estimation comprised by multilinear curve with splines was fitted at both of the experimental rooms.

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