BILINGUAL PUBLISHING CO.

01
June · 2020
Volume 2 Issue 1

# Journal of Human Physiology

Volume 2 Issue 1 · June 2020 · ISSN 2661-3859

APSCI
creative commons
CNKI 中国知网 www.cnki.net 中国知识基础设施工程
Google scholar
Crossref
MyScienceWork

ISSN 2661-3859
01
9 772661 385208
Price: S$30.00

Volume 2 Issue 1 • June 2020 • ISSN 2661-3859 (Online)

# Journal of Human Physiology



BILINGUAL PUBLISHING CO.

Pioneer of Global Academics Since 1984

BILINGUAL PUBLISHING CO.
Pioneer of Global Academics Since 1984

Volume 2 | Issue 1 | June 2020 | Page 1-33
**Journal of Human Physiology**

# Contents

## Article

BILINGUAL PUBLISHING CO.
Pioneer of Global Academics Since 1984

**Journal of Human Physiology**
https://ojs.bilpublishing.com/index.php/jhp

ARTICLE

# The Regulatory B Cell in Active Systemic Lupus Erythematosus Patients: A Systemic Review and Meta-analysis

**Xiaohuan Chen Lei Liu Lei Liao Yahui Wang Jiacheng Shi Hanyou Mo***

Department of Rheumatology, Affiliated hospital of Guilin medical university, 15 Lequn Road, Guilin, Guangxi, 541000, China

ARTICLE INFO

*Article history*
Received: 23 December 2019
Accepted: 7 February 2020
Published Online: 31 May 2020

*Keywords:*
Regulatory B cell
Systemic lupus erythematosus
Percentage
Autoimmunity
Meta-analysis

ABSTRACT

Background: The study of regulatory B cells (Bregs) in systemic lupus erythematosus (SLE) has been in full swing in recent years, but the number and function of Bregs in SLE patients have also present quite contradictory results. Therefore, we conducted a meta-analysis to verify the changes in Bregs in active SLE. Methods: We identified studies reporting the proportions of Bregs in SLE patients by searching Pubmed, Embase, Web of Science, Cochrane and CNKI. Due to the degree of heterogeneity is very high, we used a random effects model to assess the mean differences in percentages of Bregs between active SLE and controls. Then, sensitivity analysis and subgroup analysis were performed to verify potential sources of heterogeneity. Results: Seven eligible articles involving 301 active SLE patients and 218 controls were included in the meta-analysis. The pooled percentages of Bregs were found no significant difference between active SLE patients and healthy controls [0.259, (−1.150, 1.668), p = 0.719], with great heterogeneity ( $I^2$ = 97.5%) . The result of sensitivity analysis showed that exclusion of any single study or single article did not materially resolve the heterogeneity, but after excluding the article conducted by Cai X and his colleagues, the percentages of Bregs were significantly higher in active SLE than those in controls [1.394, (0.114,2.675), p = 0.033]. The results of subgroup analysis revealed that when the disease activity was judged by SLEDAI score ≥ 5, the percentages of Bregs were significantly lower in the SLE groups than in the control groups[-1.99,(-3.241,-0.739), p = 0.002], but when the threshold of SLEDAI score ≥ 6 chosen for active SLE, the percentages of Bregs were significantly increased in the SLE groups[2.546,(1.333,3.759), p < 0.001]. Meanwhile, other subgroup analysis based on the different phenotypes of Bregs, diagnostic criteria, enrolled research countries, treatment status, and organ involvement did not differ in proportion of Bregs between SLE patients and controls. Conclusions: The study implies that Bregs may play a role in the pathogenesis of active SLE, and the thresholds of SLEDAI score to distinguish between active and inactive SLE patients are important factors affecting the percentages of Bregs.

**Corresponding Author:*
*Hanyou Mo,*
*Department of Rheumatology, Affiliated hospital of Guilin medical university, 15 Lequn Road, Guilin, Guangxi, 541000, China;*
*Email: mohanyou@hotmail.com*

 DOI: https://doi.org/10.30564/jhp.v2i1.1594

## 1. Introduction

Systemic lupus erythematosus (SLE) is an autoimmune connective tissue disease involving multiple organs, which is common in young women. There are excessive activation of various immune cells and secretion of autoantibodies, and antigen-antibody complexes formed by binding of autoantibodies to antigens are deposited on skin, joints and other positions to cause tissue damage and diseases. In recent years, the role of regulatory B cells (Bregs) in SLE has been attracted widespread attention. This type cells interact with $CD4^+$ T cells by secreting cytokines , directly contacting among cells, and involving in immune responses, which are closely related to the pathogenesis and disease activity of SLE.

Bregs are a subgroup of B cells, which play a role in immune regulatory mainly through the secretion of regulatory cytokines such as interleukin(IL)-10 and transforming growth factor (TGF) –β and expression of inhibitory antibodies that inhibit pathogenic T cells and autoreactive B cells [1]. The hypothesis of regulatory B cells can suppress the immune system was first proposed in the 1970s, and they are capable of producing inhibitory antibodies to maintain this inhibitory function [2]. The regulatory function of B cells in autoimmune diseases was first discovered by Janeway et al. in experimental autoimmune encephalomyelitis (EAE) in murine. They found B cells are not required for EAE induction, but may play an immunomodulatory role in EAE from acute to complete recovery [3]. With the further research on Bregs, it has been found that there are many subtypes of Bregs, such as marginal B cells, IL-10-producing Bregs (Br1 or B10), TGF-β-producing Bregs (Br3), Foxp3-expressing Bregs and other Bregs that have cytotoxic effects [4].

Human Bregs are categorized mainly as either transitional ($CD19^+CD24^{high}CD38^{high}$) or memory ($CD24^{high}CD27^+$) [5,6]. The production of proinflammatory factors by $CD4^+$T cells can be inhibited by $CD19^+CD24^{high}CD38^{high}$ Bregs, relying on IL-10, CD80 and CD86, but not TGF-β [6]. $CD19^+CD25^{high}CD86^{high}CD1d^{high}$ Bregs produce IL-10 and TGF-β that inhibit $CD4^+$T cells proliferation and enhance expression of FoxP3 and T lyphocyte antigen 4 in regulatory T cells [7]. $CD24^{high}CD27^+$ Bregs also regulate the production of TNF-α by producing IL-10 [6]. Therefore, human Bregs are not a single phenotype, but regardless of phenotype, most of their regulatory function depends on IL-10. Although there are few studies on human regulatory B cells, evidence suggested that they may become targets for the treatment of human immune diseases in the future.

Despite these evidences, we still lack confidence in the beneficial effects that therapeutic Bregs may have on SLE patients. The use of Breg-based therapies should be based on changes in the number of Bregs and/or impaired regulatory function associated with the pathogenesis of SLE. However, the results of studies on the number of Bregs in active SLE patients and normal healthy people are quite contradictory; the frequency of Bregs in SLE patients is reduced or increased [8-14]. Importantly, the role of Bregs in SLE is also controversial. It is conceivable that quantifying Breg's strategy is crucial to draw conclusions about Breg subtypes. In addition, differences in patients recruitment (research country, diagnostic criteria, treatment status, disease activity, organ involvement) may also be the cause of significant differences in the literature. However, to the best of our knowledge, no source of these inconsistent results has been studied. It is still unclear that the quantitative and qualitative changes about Bregs in SLE, but immunotherapy based on Bregs shows promising therapeutic power, thus we performed this meta-analysis to obtain more information about Bregs in SLE patients, explore the reasons for inconsistent sources of results, and gain a more detailed understanding of the role of Bregs in the pathogenesis of SLE.

## 2. Methods

### 2.1 Search Strategy

The literature search was conducted in Pubmed, Embase, Web of Science, Cochrane and CNKI using the MeSH terms "regulatory B cell" and "systemic lupus erythematosus" and their combination. We searched for relevant studies that were updated to October 20, 2019. All potentially eligible articles were also considered except for murine experiments, reviews and conference abstract superseded by publication. There were no limits on geographical location and ethnicity.

### 2.2 Eligibility Criteria

Studies that fulfilled the following criteria were included: (1) evaluating the levels of Bregs in SLE patients and controls; (2)the levels of Bregs were presented as ratio of Bregs to total lymphocytes(%); (3) available as a full text article; (4) providing mean (standard deviation/ standard error) or median (range/ interquartile range); (5) case-control study. Reviews, studies about murine experiments, conference abstracts that were not published as full-length articles were not included.

### 2.3 Data Extraction

Two independent researchers selected and recorded eligible articles. The other researchers were consulted to reach a consensus when any divergence occurred. The following



DOI: https://doi.org/10.30564/jhp.v2i1.1594

information was extracted from the studies: first author's name, publication time, regions where the authors performed studies, diagnostic criteria, Breg definition, treatment status, threshold of SLEDAI chosen to define active SLE, the number of patients and controls, the frequency of Bregs(%). When the medians and ranges (or interquartile ranges) were provided in studies instead of means and standard deviations, we calculated the means and standard deviations by estimation methods [15]. The quality of included studies was evaluated by the Newcastle-Ottawa Quality Assessment Scale (NOS).

### 2.4 Statistical Analysis

We used the $I^2$-statistic to explore the heterogeneity among studies. The $I^2$ values of 25, 50, and 75%, respectively, were used as evidence of low, medium, and high heterogeneity. When the pooled results were in high heterogeneity, a random effects model was used, and a fixed-effects model was used in the case of low heterogeneity or no heterogeneity. We performed other analysis when heterogeneity was high, including subgroup analysis and publication bias to explore heterogeneity. We explored the heterogeneity of researches by drawing forest plots to visualize the results more intuitively. By examining funnel plot asymmetry using the Begger and Egger tests ($p \geq 0.05$), we assessed the publication bias. We also performed a sensitivity analysis to test the robustness of the original results. All statistical analysis were performed using Stata software (ver.15.0). This meta-analysis was conducted according to the PRISMA guidelines.

## 3. Results

### 3.1 Literature Search

We searched the database for 1779 articles that might be eligible. The flow chart about the screening process of literatures is shown in Figure 1. We excluded 1677 articles by screening headlines and abstracts. Then 16 duplicated articles were excluded, 15 articles were not case-control trials, 15 articles were not for SLE patients, 14 articles were conference abstract superseded by publications, 17 articles did not provide relevant data, and 18 articles could not obtain full-text information. Therefore, this meta-analysis included a total of seven articles [8-14].

### 3.2 Study Characteristics

All main characteristics of the included studies are presented in Table 1. These studies were published between 2014 and 2019. The analysis included 301 active SLE patients and 218 controls from seven eligible articles. Of these articles, five were conducted in China [8,10,12-14], one in Israel [11] and one in Germany [9]. The diagnose criteria of SLE varied across studies. All controls were healthy people without any autoimmune disease. We regarded all studies as case-control studies and scored them using the NOS, and all studies had a scored of 5-6.

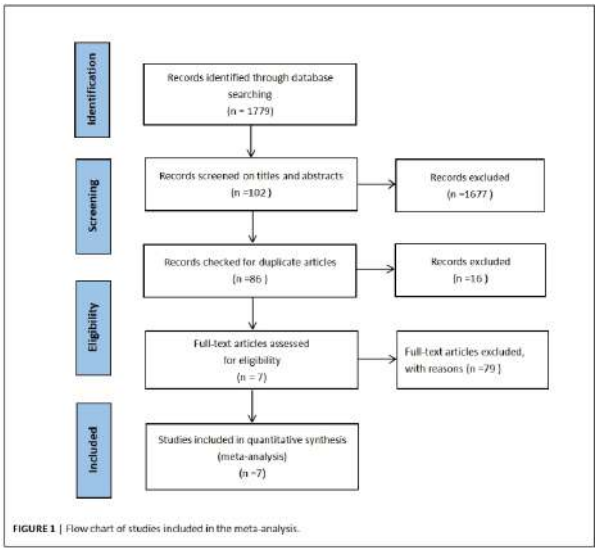

FIGURE 1 | Flow chart of studies included in the meta-analysis.

**Table 1.** Characteristics of studies included in the meta-analysis

| References | Region | Diagnosis criteria | Treatment status | Threshold of SLEDAI for active SLE | Breg definition | Case (n) | Control (n) | Bregs in case (mean±SD,%) | Bregs in control (mean±SD,%) | NOS score |
|---|---|---|---|---|---|---|---|---|---|---|
| Cai X et al. | China | 1997 | Not report | ≥5 | IL-10+CD19+ | 38 | 20 | 1.54±0.64 | 4.35±1.00 | 6 |
| | | | | | CD19+CD24highCD38high | 38 | 20 | 1.26±0.45 | 3.14±0.87 | |
| Heinemann K et al. | Germany | *ACR | Partial treated | >4 | IL-10+ | 34 | 21 | 5.50±4.80 | 17.00±7.00 | 5 |
| | | | | | CD19+CD24highCD38high | 34 | 21 | 1.60±2.60 | 1.50±1.10 | |
| Wang H et al. | China | 1982 | Partial treated | >5 | CD19+CD24highCD38high | 30 | 30 | 2.70±1.97 | 0.38±0.33 | 5 |
| Vadasz Z et al. | Israel | 1992 | Untreated | Not report | CD19+CD25high | 21 | 20 | 18.50±3.05 | 11.00±1.65 | 5 |
| Wang T et al. | China | 1997 | Not report | Not report | CD19+CD24highCD38high | 56 | 35 | 39.83±21.39 | 8.74±3.97 | 5 |
| Yang X et al. | China | 1997 | Treated | ≥6 | CD19+CD5+CD1dhigh | 16 | 15 | 4.90±1.27 | 1.63±0.99 | 5 |
| | | | | | IL-10+CD19+ | 6 | 6 | 3.44±0.69 | 1.15±0.45 | |
| Wang Z et al. | China | 1997 | Untreated | ≥5 | CD19+CD24highCD38high | 28 | 30 | 2.10±1.09 | 4.07±1.48 | 5 |

***Notes:*** *ACR without detailed description

### 3.3 Meta-Analysis of the Breg Percentages in Active SLE Patients and Publication Bias

Initially, we compared the percentages of Bregs in active SLE patients and healthy controls, regardless of the Breg definitions were used. A total of ten studies were available in seven eligible articles, and five studies reported higher percentage of Bregs in active SLE patients than those in the control group [10-13], three studies reported decreased percentages [8,14]. In addition, the simultaneous analysis of two different subtypes of Breg in two studies from one articles yielded conflicting results [9]. Surprisingly, in the overall analysis, there was no significant difference in any studies [0.259, (−1.150, 1.668), p = 0.719, Figure 2]. Meanwhile, the heterogeneity was 97.5% (p < 0.001) by $I^2$ statistic and thus very high, and a random effect model was used for meta-analysis. The Egger test showed no publication bias (t = 0.13, p = 0.898, Figure 3).

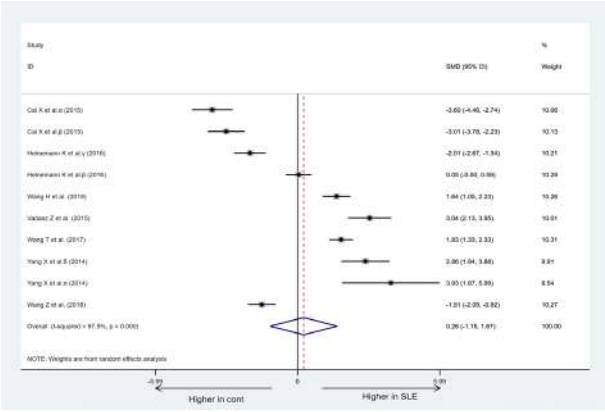


**Figure 2.** Forest plot of the percentage changes of Bregs in active SLE patients compared with the controls. α: Bregs were gated by IL-10$^+$CD19$^+$; β: Bregs were gated by CD19$^+$CD24$^{high}$CD38$^{high}$; γ: Bregs were gated by IL-10$^+$; δ: Bregs were gated by CD19$^+$CD5$^+$CD1d$^{high}$

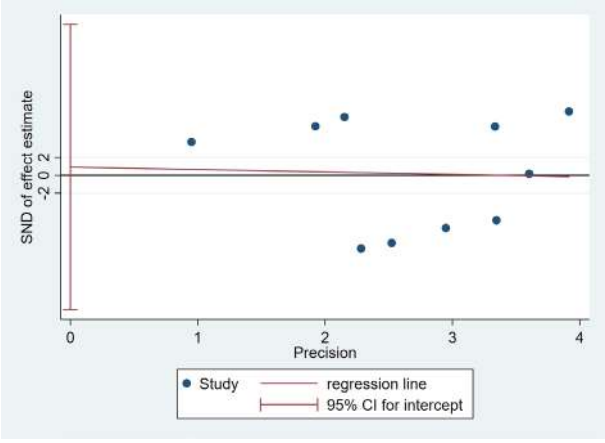


**Figure 3.** Publication bias analysis using Egger linear regression and Begg rank correlation test

### 3.4 Sensitivity Analysis and Subgroup Analysis

Due to the high heterogeneity of the results, we performed a sensitivity analysis. Then, considering the different phenotypes of Bregs, diagnostic criteria, enrolled regions, threshold of SLEDAI chosen for active SLE definition, treatment status, and organ involvement are potential factors that may lead to bias in the results, we performed a subgroup analysis based on these factors.

Firstly, we performed sensitivity analysis to explore potential sources of heterogeneity. Exclusion of any single study or single article did not materially resolve the heterogeneity. But after excluding the article conducted by Cai X and his colleagues [8], the percentages of Bregs were significantly higher in active SLE than those in controls [1.394, (0.114,2.675), p = 0.033].

Then, we hypothesized that the main cause of unexpected results may be the inconsistent definition of Bregs. Therefore, we performed a subgroup analysis based on the Bregs definition to explore potential sources of heterogeneity. There are several subtypes in the selected articles, which can roughly divide Bregs into two groups: based on IL-10$^+$ and CD19$^+$(Table 2). To our surprise, no matter whether Breg was based on IL-10$^+$ or CD19$^+$, there was no statistical difference between SLE patients and control groups. When Bregs were gated based on IL-10$^+$, the percentages of Bregs in active SLE were comparable to those in the controls [-0.783,(-3.526,1.960), p = 0.576, Figure 4] [8,9,13]. Meanwhile, pooled analysis of nine studies revealed the proportion of Bregs defined as CD19$^+$ cells did not differ significantly between active SLE patients and healthy controls [0.517,(-0.979,2.014), p = 0.498, Figure 5] [8,10-14].

**Table 2.** Subgroup analysis based on different definitions of Bregs in patients with SLE

| Definition of Bregs | Number of studies | Test of association | | |
|---|---|---|---|---|
| | | SMD | 95%CI | P value |
| Association with IL-10-positive | 3 | -0.783 | (-3.526,1.960) | 0.576 |
| IL-10+CD19+ | 2 | 0.106 | (-7.272,7.484) | 0.978 |
| IL-10+ | 1 | -2.007 | (-2.672,-1.342) | - |
| Association with CD19-positive | 9 | 0.517 | (-0.979,2.014) | 0.498 |
| IL-10+CD19+ | 2 | 0.106 | (-7.272,7.484) | 0.978 |
| CD19+CD24highCD38high | 5 | -0.186 | (-1.856,1.483) | 0.827 |
| CD19+CD5+CD1dhigh | 1 | 2.86 | (1.841,3.879) | - |
| CD19+CD25high | 1 | 3.037 | (2.127,3.948) | - |

From the articles we collected, it was found that the researchers did not agree on the diagnostic criteria for active SLE patients. Six of the studies were based on the 1997 diagnostic criteria [8,12-14], one based on the 1982 ACR standard [10], and three studies showing only the ACR criteria without specific years [9,11]. Therefore, we conducted



DOI: https://doi.org/10.30564/jhp.v2i1.1594

a subgroup analysis to explore whether it is a source of heterogeneity. However, our subgroup analysis showed no statistically significant difference between the percentages of Bregs calculated using the 1997 diagnostic criteria and the ACR diagnosis criteria [0.008,(-2.203,2.219), p = 0.994 for subgroup of 1997, 0.340,(-2.137,2.817), p = 0.788 for subgroup of ACR, Figure 6].

More and more recent studies have shown that SLE has geographical and ethnic differences [16], so we assumed that the heterogeneity was caused by the different countries of the research population. There are differences in the prevalence and incidence of SLE in different countries, which may have a certain impact on the percentages of Bregs. Of the ten studies, seven were conducted in China [8,10,12-14], two in Germany [9], and one in Israel [11]. However, by subgroup analysis, the percentages of Bregs in each subgroup of SLE patients and controls was still not statistically significant(Figure 7).

Our results revealed that choosing the SLEDAI score to distinguish thresholds between active SLE and inactive SLE may result in heterogeneity. We attributed the study with a SLEDAI score of >4 to the ≥5 group [9], and the study with a score of >5 to the ≥6 group [10]. Therefore, ultimately all studies could be divided into three subgroups, "≥5" group, "≥6"group and "not reported" group(Figure 8). When the disease activity was judged by SLEDAI score ≥ 5, the percentages of Bregs were significantly lower in the SLE groups than in the control groups[-1.99,(-3.241,-0.739), p = 0.002] [8,9,14], but when the threshold of SLEDAI score ≥ 6 chosen for active SLE, the percentages of Bregs were significantly increased in the SLE groups[2.546,(1.333,3.759), p < 0.001] [10,13].

Previous study suggested that glucocorticoid therapy can increase the frequency of Bregs in patients with SLE [17]. The subgroup treatment status analysis of this meta-analysis showed no statistically significant differences among studies receiving drug therapy or those partially receiving drugs or no drug therapy(Figure 9). Given the fact that most patients who enrolled in the current studies received medication and the differences in the use of drugs, the possible effects of the treatment require further evaluation.

SLE can involve many organs, and lupus nephritis (LN) is a typical major organ involving of SLE. In all articles, Heinemann K et al. [9] compared the percentages of two different Breg subtypes in LN with the healthy control group. The data showed that the percentages of Bregs in active LN was not statistically different from the healthy controls[-1.010,(-2.882, 0.863), p < 0.291].

To assess the impact of disease activity further, we analyzed the percentages of Breg in active SLE versus in inactive SLE and in inactive SLE versus in healthy controls. The results presented are also not statistically significant.

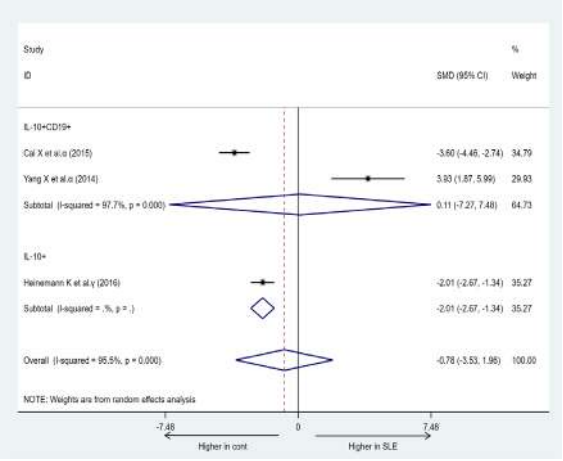


**Figure 4.** Subgroup analysis based on IL-10$^+$ as definition of Bregs in SLE patients

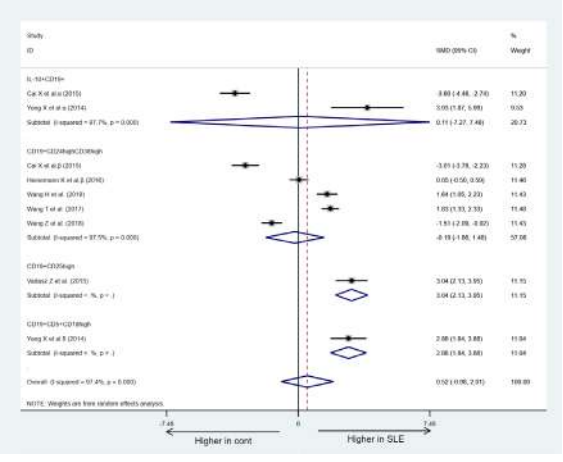


**Figure 5.** Subgroup analysis based on CD19$^+$ as definition of Bregs in SLE patients

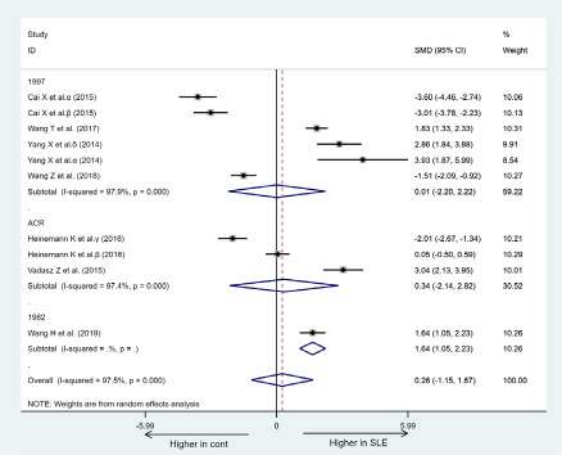


**Figure 6.** Subgroup analysis based on different diagnose criterion in SLE patients

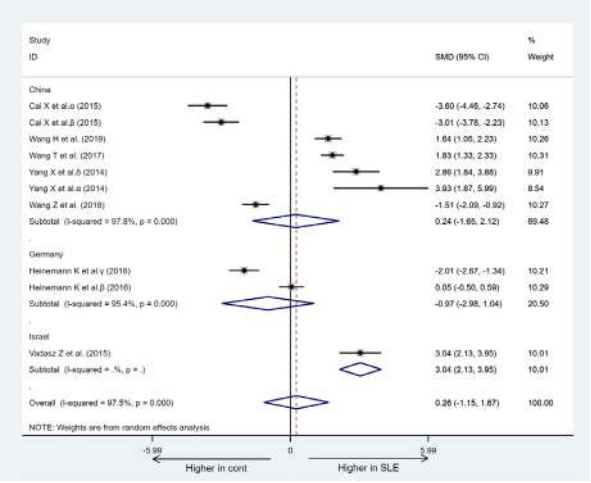


**Figure 7.** Subgroup analysis based on different study regions in SLE patients

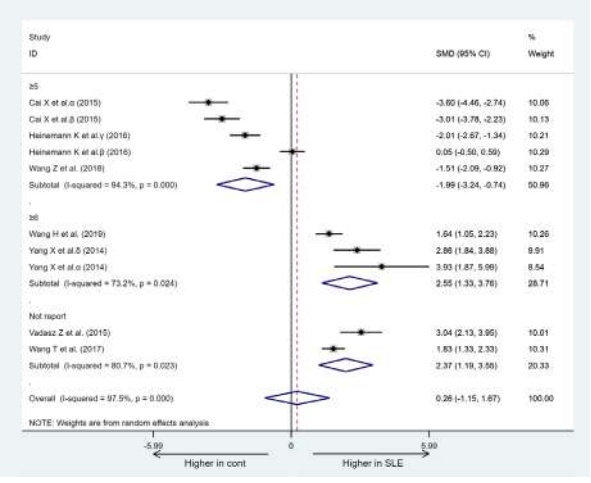


**Figure 8.** Subgroup analysis based on different thresholds of SELDAI chosen for active SLE definition in SLE patients

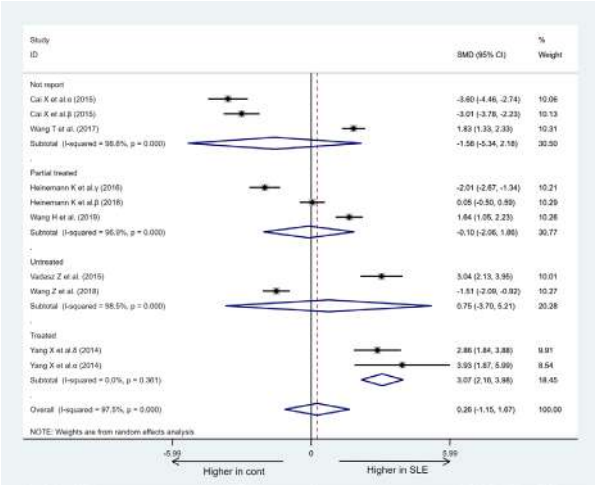


**Figure 9.** Subgroup analysis based on different treatment status in SLE patients

## 4. Discussion

B cells are one of the main components of specific immunity, and play a positive regulatory role mainly by initiating and activating immunity. Including secretion of antibodies, antigen presentation, production of inflammation and immunoregulatory factors, providing synergistic stimulation signals, etc. It is known that T cells have immunoregulatory functions, and whether B cells have regulatory subpopulations has become a research hotspot in recent years. Increased or decreased percentage of Bregs has been reported in patients with active SLE [8-14]. Therefore, we conducted further research on these results. However, meta-analysis results found no significant difference in percentages of Bregs between active SLE patients and healthy controls. Since the results showed great heterogeneity ($I^2$> 90), sensitivity analysis and subgroup analysis was subsequently performed to evaluate the possible effect of several factors on the percentages of Bregs, and the results indicated that different thresholds of SLEDAI chosen for defining active SLE might influence the frequency.

When studying Bregs, it is important to determine their phenotype. Previous research found that Bregs can produce IL-10, TGF-β, and Foxp3. One subtype of Bregs mainly secrete IL-10, which can inhibit the differentiation of T helper cells 1 (Th1) and T helper cells 17 (Th17) , and reduce the production of inflammatory cytokines through dendritic cells [18]. Such IL-10-producing Bregs are called B10 cells. In humans, B10 cells account for less than 1% of the total number of B cells in the blood. Human Bregs include $CD19^{+}CD24^{high}CD38^{high}CD1d^{high}$ and $CD19^{+}CD24^{high}CD27^{+}$, etc [19]. The relationship between development and differentiation of these subtypes of Bregs is unclear. In addition, Breg functioning involves CD40, TLR, B cell receptor, CD19, CD1d, etc [20]. In this study, no statistically significant difference was found in the different statistical strategies of Bregs subtype classification. Normal frequencies were observed when $IL\text{-}10^{+}$ was chosen to define Bregs [-0.783, (-3.526,1.960), p = 0.576] [8,9,13]. The percentages of Breg based on $CD19^{+}$ did not show a significant difference between the patients and the control group [0.517 , (-0.979, 2.014), p = 0.498] [8,10-14]. Considering that some studies only mentioned $IL\text{-}10^{+}$ Bregs, but did not mention the specific Bregs subtypes that secreted IL-10, and the research sample size was insufficient, further researches on the frequency of Bregs with different subtypes are needed in the future.

Since SLE is a heterogeneous disease, the diagnosis is not always so simple. From American College of Rheumatology (ARA) established the SLE classification standard in 1971 [21], many changes have taken place in the diag-



DOI: https://doi.org/10.30564/jhp.v2i1.1594

nostic criteria for SLE. The effect of different diagnostic criterion on the percentages of Bregs was not found in our study, and the results showed no statistical significance. However, the underlying causes and mechanisms leading to this result need to be further clarified in the future.

Recently, many studies have shown that there are significant regional differences in the incidence, clinical manifestations, and mortality of SLE. Khan A et al. [22] showed that significant regional differences existed in the clinical manifestations of SLE in Khyber Pakhtunkhwa compared to other regions through cross-sectional studies. In addition, Yen EY et al. [23] found that the United States since 1968, mortality of SLE has declined, but it remains higher than mortality in non-SLE, and there were significant gender, ethnic, and regional differences. Therefore, we hypothesized that differences in the study regions may result in variability in the results, but unexpectedly, the analysis results showed that the percentages of Breg in SLE patients from different regions was comparable to these of the healthy control group. We considered that most of researches originated in China, while the sample size of other countries was small, China had a large land area, a large span of latitudes and longitudes, different environments and ethnicities, which would also cause heterogeneity of Bregs in SLE patients. Thus, a larger sample size studies are needed for analysis.

The SLEDAI score can be used to judge the condition of SLE. Different scores indicate differences in disease activity, which determine the use of different doses of glucocorticoid and the choice of different immunosuppressants [24]. Our results revealed that active SLE could cause heterogeneity by choosing different thresholds of SLEDAI score. When SLEDAI score $\geq 5$ was defined as active SLE, the percentages of Bregs in patients with active SLE were significantly lower than those in the healthy control group [-1.99, (-3.241-0.739), $p = 0.02$] [8,9,14]; however, the percentages of Bregs in patients with active SLE were significantly higher than control groups, when SLEDAI score $\geq 6$ as the activity standard [2.546, (1.333, 3.759), $p < 0.001$] [10,13]. Considering that higher thresholds of SLEDAI score in recruited SLE patients may indicate a more severe conditions, Bregs seems to be positively correlated with disease activity in SLE, but this does not explain that the percentages of Bregs in SLE patients with a slightly lower SLEDAI score were lower than in healthy controls. Therefore, the changes of Bregs in the process of SLE disease need to be further studied.

Drug treatment for patients with SLE includes glucocorticoid, hydroxychloroquine, cyclophosphamide, and azathioprine etc. Previous studies showed that glucocorticoid therapy can increase the frequency of Bregs in patients with SLE [17]. However, the use of cyclophosphamide appeared to reduce the amount of Bregs [25]. Results of the subgroup analysis based on treatment status did not reveal a statistical difference between patients who received drug therapy and untreated patients. Drugs for SLE are diverse, and each SLE patient enrolled in the studies received different treatment status and used different drugs. Thus the possible effects of different treatment modalities on Bregs need to be further evaluated in the future.

Recently, studies by Szabó K et al. and Makiyama A. et al. found that activated T helper cells, especially Th1 cells, were associated with the promotion of B cell differentiation in SLE patients [26-27]. On the one hand, Bregs can secrete IL-10, TGF-β and other related factors, inhibit the conversion of Th0 to Th1 and Th2, reduce the production of inflammatory factors such as TNF-α and IFN-γ, and down-regulate autoimmune response and excessive immunity [28-29]; On the other hand, Bregs can exert immunoregulatory effects by affecting the number and function of Tregs [30]; Bregs can also suppress the immune response through cell-to-cell contact, such as by contacting CD40 / CD40L with effector T cells and causing T cells death [31]. It seems that the Bregs and T cells can interact and influence each other. In the past, Zhang SX et al. [32] systematically reviewed and performed a meta-analysis about the proportion of Tregs in patients with SLE, the results showed that the percentages of Tregs in SLE patients was significantly lower than those in healthy controls. Considering the correlation between Bregs and Tregs, we evaluated the proportion of Bregs in SLE patients, and analyzed the relevant influencing factors to obtain more evidence about Bregs in the pathogenesis of SLE, which may aid in the development of approaches to the therapeutic modalities employing the use of Bregs.

The present study has certain limitations. First of all, not all research-related information is publically available, such as the duration of the disease, specific information on the use of drugs or treatments, etc., and we did not contact the corresponding authors in time to obtain more information, which prevented us from comprehensive investigation of the disease course and the effect of drugs on the percentages of Bregs. Second, our assessment of the factors affecting the percentages of Bregs was inadequate. Faced with a high degree of heterogeneity, more SLE patients who are in different backgrounds and disease status need to be studied, which may help to clarify the role and changes of Bregs in SLE disease process better.

## 5. Conclusion

In summary, our meta-analysis results suggest that Bregs may play a role in the pathogenesis of active SLE, and the

 DOI: https://doi.org/10.30564/jhp.v2i1.1594

thresholds of SLEDAI score to distinguish whether SLE patients are active or not are important factors affecting the percentages of Bregs. Our findings support the notion that Bregs status is important in patients with SLE, but we cannot determine how Bregs have changed throughout the course of SLE. Due to the evidence is still limited, more and further large-scale and well-designed randomized controlled trials are urgently needed.

## Author Contributions

MHY: study design. LL and LL: data collection. CXH, WYH and SJC: statistical analysis. CXH: paper writing. MHY: paper revision. All authors approved the submitted version of the manuscript.

## Funding

This work was supported by funding from the National Natural Science Foundation of China Grants [81760286].

## References

[1] Mizoguchi A, Bhan AK. A case for regulatory B cells[J]. J Immunol, 2006, 176: 705-710.
DOI: 10.4049/jimmunol.176.2.705

[2] Morris A, Moller G. Regulation of cellular antibody synthesis effect of adoptively transferred antibody-producing spleen cells on cellular antibody synthesis[J]. J Immunol, 1968, 101: 439-445.
DOI: 10.1084/jem.127.2.291

[3] Wolf SD, Dittel BN, Hardardottir F, et al. Experimental autoimmune encephalomyelitis induction in genetically B cell-deficient mice[J]. J Exp Med, 1996, 184: 2271-2278.
DOI: 10.1084/jem.184.6.2271

[4] Berthelot JM, Jamin C, Amrouche K, et al. Regulatory B cells play a key role in immune system balance[J]. Joint Bone Spine, 2013, 80: 18-22.
DOI: 10.1016/j.jbspin.2012.04.010

[5] Blair PA□Norena LY□Flores-Borja F, et al. CD19 (+) CD24 (hi)CD38(hi) B cells exhibit regulatory capacity in healthy individuals but are functionally impaired in systemic Lupus Erythematosus patients[J]. Immunity, 2010, 32(1) : 129-140.
DOI: 10.1016/j.immuni.2009.11.009

[6] Iwata Y, Matsushita T, Horikawa M, et al. Characterization of a rare IL-10-competent B-cell subset in humans that parallels mouse regulatory B10 cells[J]. Blood, 2011, 117(2): 530-541.
DOI: 10.1182/blood-2010-07-294249

[7] Kessel A, Haj T, Peri R, et al. Human CD19 (+) CD25 (high) B regulatory cells suppress proliferation of CD4 (+) T cells and enhance Foxp3 and CTLA-4 expression in T-regulatory cells[J]. Autoimmunity Rev, 2012, 11(9):670-677.
DOI:10.1016/j.autrev.2011.11.018

[8] X Cai, X Li, X Lin, et al. Expression of regulatory B cells in peripheral blood of patients with systemic lupus erythematosus[J]. Zhonghua Yi Xue Za Zhi. 2015, 95(17): 1310-3.
DOI:10.3760/cma. j. issn. 0376-2491. 2015. 17. 007

[9] Heinemann K, Wilde B, Hoerning A, et al. Decreased IL-10(+) regulatory B cells (Bregs) in lupus nephritis patients[J]. Scand J Rheumatol. 2016, 45(4): 312-6.
DOI:10.3109/03009742.2015.1126346

[10] H Wang, C Liu, W Chen, G Ding. The skewed frequency of B-cell subpopulation CD19+CD24hiCD38hi cells in peripheral blood mononuclear cells is correlated with the elevated serum sCD40L in patients with active systemic lupus erythematosus[J]. J Cell Biochem. 2019: 1-8.
DOI:10.1002/jcb.28427

[11] Vadasz Z, Peri R, Eiza N, Slobodin G, Balbir-Gurman A, Toubi E. The Expansion of CD25 high IL-10 high FoxP3 high B Regulatory Cells Is in Association with SLE Disease Activity[J]. J Immunol Res. 2015, 2015: 254245.
DOI: 10.1155/2015/254245

[12] T Wang, Z Li, X Li, et al. Expression of CD19+CD24highCD38high B cells, IL-10 and IL-10R in peripheral blood from patients with systemic lupus erythematosus[J]. Mol Med Rep. 2017, 16(5): 6326-6333.
DOI: 10.3892/mmr.2017.7381

[13] X Yang, J Yang, Y Chu, et al. T follicular helper cells and regulatory B cells dynamics in systemic lupus erythematosus[J]. PLoS One. 2014, 9(2): e88441.
DOI: 10.1371/journal.pone.0088441

[14] Z Wang, F Wang, Y Wang. Study on CD19+CD24hiCD38hiBreg cells in the patients with systemic lupus erythematosus[J]. Chinese modern doctor. 2018, 56(32): 15-18.
DOI: CNKI:SUN:ZDYS.0.2018-32-006

[15] Hozo SP, Djulbegovic B, Hozo I. Estimating the mean and variance from the median, range, and the size of a sample[J]. BMC Med Res Methodol. 2005, 5: 13.
DOI: 10.1186/1471-2288-5-13

[16] Rees F, Doherty M, Grainge MJ, Lanyon P, Zhang W. The worldwide incidence and prevalence of systemic lupus erythematosus: a systematic review of epidemiological studies[J]. Rheumatology (Oxford). 2017, 56(11): 1945-1961.
DOI: 10.1093/rheumatology/kex260

[17] XM Deng, SX Yan, W Wei. The role of regulatory

  DOI: https://doi.org/10.30564/jhp.v2i1.1594

B cells in SLE and the effect of glucocorticoids[C]. Collection of papers of Anhui pharmacology society annual meeting, 2012, 2012: 29-30.
http://kns.cnki.net/kcms/detail/detail.aspx?dbcode=CPFD&filename=AHSK201209002033&dbname=CPFD0914

[18] Matsumoto M, Baba A, Yokota T, et al. Interleukin-10-producing plasmablasts exert regulatory function in autoimmune inflammation[J]. Immunity. 2014, 41(6): 1040-51.
DOI: 10.1016/j.immuni.2014.10.016

[19] Iwata Y, Matsushita T, Horikawa M, et al. Characterization of a rare IL-10-competent B-cell subset in humans that parallels mouse regulatory B10 cells[J]. Blood. 2011, 117(2): 530-41.
DOI: 10.1182/blood-2010-07-294249

[20] Rincón-Arévalo H, Sanchez-Parra CC, Castaño D, Yassin L, Vásquez G. Regulatory B Cells and Mechanisms[J]. Int Rev Immunol. 2016, 35(2): 156-76.
DOI: 10.3109/08830185.2015.1015719

[21] Gibson TP, Dibona GF. Use of the American Rheumatism Association's preliminary criteria for the classification of systemic lupus erythematosus. Ann Intern Med. 1972, 77(5): 754-6.
DOI: 10.7326/0003-4819-77-5-754

[22] Khan A, Shah MH, Nauman M, et al. Clinical manifestations of patients with Systemic Lupus Erythematosus (SLE) in Khyber Pakhtunkhwa[J]. J Pak Med Assoc. 2017, 67(8): 1180-1185.
https://jpma.org.pk/article-details/8308?article_id=8308

[23] Yen EY, Shaheen M, Woo J, et al. 46-Year Trends in Systemic Lupus Erythematosus Mortality in the United States, 1968 to 2013: A Nationwide Population-Based Study[J]. Ann Intern Med. 2017, 167(11): 777-785.
DOI: 10.7326/M17-0102

[24] Yee CS, Farewell VT, Isenberg DA, et al. The use of Systemic Lupus Erythematosus Disease Activity Index-2000 to define active disease and minimal clinically meaningful change based on data from a large cohort of systemic lupus erythematosus patients[J]. Rheumatology (Oxford). 2011, 50(5): 982-8.
DOI: 10.1093/rheumatology/keq376

[25] J Fan, J Luo, C Yan, et al. Methotrexate, combined with cyclophosphamide attenuates murine collagen induced arthritis by modulating the expression level of Breg and DCs[J]. Mol Immunol. 2017, 90: 106-117.
DOI: 10.1016/j.molimm.2017.07.001

[26] Szabó K, Papp G, Szántó A, Tarr T, Zeher M. A comprehensive investigation on the distribution of circulating follicular T helper cells and B cell subsets in primary Sjögren's syndrome and systemic lupus erythematosus[J]. Clin Exp Immunol. 2016, 183(1): 76-89.
DOI: 10.1111/cei.12703

[27] Makiyama A, Chiba A, Noto D, et al. Expanded circulating peripheral helper T cells in systemic lupus erythematosus: association with disease activity and B cell differentiation[J]. Rheumatology (Oxford). 2019, 58(10): 1861-1869.
DOI: 10.1093/rheumatology/kez077

[28] Iwata Y, Matsushita T, Horikawa M, et al. Characterization of a rare IL-10-competent B-cell subset in humans that parallels mouse regulatory B10 cells[J]. Blood. 2011, 117(2): 530-41.
DOI: 10.1182/blood-2010-07-294249

[29] Tedder TF. B10 cells: a functionally defined regulatory B cell subset[J]. J Immunol. 2015, 194(4): 1395-401.
DOI: 10.4049/jimmunol.1401329

[30] Ray A, Basu S, Williams CB, Salzman NH, Dittel BN. A novel IL-10-independent regulatory role for B cells in suppressing autoimmunity by maintenance of regulatory T cells via GITR ligand[J]. J Immunol. 2012, 188(7): 3188-98.
DOI: 10.4049/jimmunol.1103354

[31] N Gao, Dresel J, Eckstein V, et al. Impaired suppressive capacity of activation-induced regulatory B cells in systemic lupus erythematosus[J]. Arthritis Rheumatol. 2014, 66(10): 2849-61.
DOI: 10.1002/art.38742

[32] SX Zhang, XW Ma, YF Li, et al. The Proportion of Regulatory T Cells in Patients with Systemic Lupus Erythematosus: A Meta-Analysis[J]. J Immunol Res. 2018, 2018: 7103219.
DOI: 10.1155/2018/7103219

 DOI: https://doi.org/10.30564/jhp.v2i1.1594

BILINGUAL PUBLISHING CO.
Pioneer of Global Academics Since 1984

**Journal of Human Physiology**
https://ojs.bilpublishing.com/index.php/jhp

ARTICLE

# The Use of Electromagnetic Forces of the Earth in Manual and Physiotherapy

**Yuri Pivovarenko***

Research and Training Center "Physical and Chemical Materials Science" Under Kyiv Taras Shevchenko University and NAS of Ukraine, Kiev, Ukraine

ARTICLE INFO

*Article history*
Received: 27 December 2019
Accepted: 10 February 2020
Published Online: 31 May 2020

*Keywords:*
Manual therapy
Physiotherapy
Electrets
Yoga

ABSTRACT

Physiotherapists usually ignore the electric polarization of human bodies that occurs under the influence of the electromagnetic forces of the Earth. This is irrational, since the positive or negative electrification of human tissues has the opposite effect on both their properties and functional activity. How physiotherapists must take into account the polarizing effect of the electromagnetic forces of the Earth when analysing the functional states of the tissues of the human body is shown here. It also shows how these electromagnetic forces can be used by manual and physiotherapists.

## 1. Introduction

Electromagnetic forces of understandable origin constantly act on the earth's surface. First of all, such forces include the Lorentz forces arising from the diurnal rotation of the Earth relative to the geomagnetic field (Figure 1).

Thus, thanks to the daily rotation of the Earth, its atmosphere and surface intersect the horizontal lines of the geomagnetic field. Therefore, both objects of the earth's surface and objects of the earth's atmosphere are constantly exposed to the Lorentz force $F_L$, which is directed upward and therefore moves positive charges up and negative charges down. The fact that this force is significant is confirmed by calculations according to which this force is capable of accelerating up a single proton up to $4{,}175 \cdot 10^7$

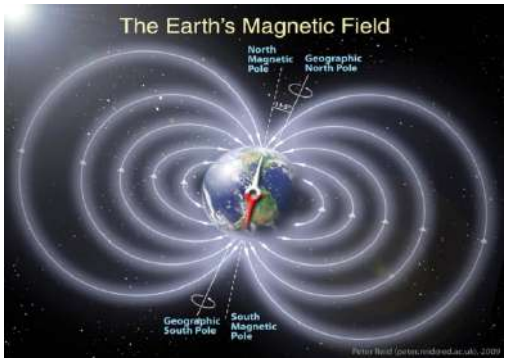


**Figure 1.** With the Earth's daily rotation, all terrestrial objects constantly cross the lines of force of the geomagnetic field. For this reason, they are constantly affected by the Lorentz force $F_L$, directed upward, and the Lorentz force $F_L^*$, directed to the equator

**Corresponding Author:*
*Yuri Pivovarenko,*
*Research and Training Center "Physical and Chemical Materials Science" Under Kyiv Taras Shevchenko University and NAS of Ukraine, Kiev, Ukraine;*
*Email: y.pivovarenko@gmail.com*



DOI: https://doi.org/10.30564/jhp.v2i1.1608

$m \cdot s^{-2}$, and a single electron to $2{,}7 \cdot 10^{12}$ $m \cdot s^{-2}$, in any case, on the equator line. In addition, calculations show that under the influence of the same Lorentz force $F_L$, one proton can "raise" a drop consisting of ~ 84830 water molecules ($\sim 1{,}5 \cdot 10^6$ D) [1,2].

Given the results of such calculations, it is not surprising that this Lorenz force $F_L$ is able to quickly and efficiently distribute positively and negatively charged water vapor in the clouds (Figure 2) [3].

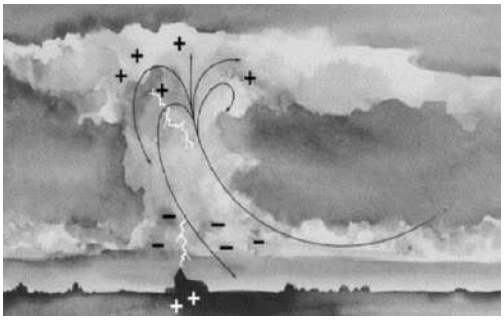

**Figure 2.** Polarization of clouds: the lower part of a typical cloud has a negative charge and the upper part has a positive charge [2,3]

All this makes it possible to assume that the same Lorentz force $F_L$ causes a positive charge of the upper atmosphere and a negative charge on the Earth's surface. The fact that such a distributive effect of the Lorentz force $F_L$ is significant is confirmed by the fact that the upper layers of the earth's atmosphere have a potential of ~ $1 \cdot 10^5$ V relative to the earth's surface [3].

As well, it is not also surprising that the same Lorenz force $F_L$ is able to create the celestial discharges, both up directed positively charged (Figure 3, left), and down directed negatively charged (Figure 3, right) [2].

**Figure 3.** Left: these are blue jets representing ascending currents of hydrated protons. Right: these are ordinary thunderstorms, which are downward flows of hydrated electrons [2]

Given all this, it is not surprising that this Lorentz force $F_L$ can cause electric polarization of the body of a standing or sitting person. Thus, when a person is standing or sitting, his head is positively charged, and his legs are negatively charged, like the cloud in Figure 2.

There is also a polarization effect of the Lorentz force $F_L^*$, arising due to the interaction of the moving surface of the Earth with the vertical component of the geomagnetic field (Figure 1) and directed towards the equator. Thus, the head and legs of a person lying along the meridian turn out to be charged differently.

The effect of both of these polarizations on the human body is discussed here. The possible medical use of these polarizations is also being discussed.

## 2. Results

***Properties of Water, Depending on its Electrical Potential***

It was discovered that the electrical potential of the water determines its ability to hydrate some polymers of biological origin. In particular, it has been shown that water with a positive potential better hydrates biological polymers than water with a negative potential (Figure 4) [4, 5].

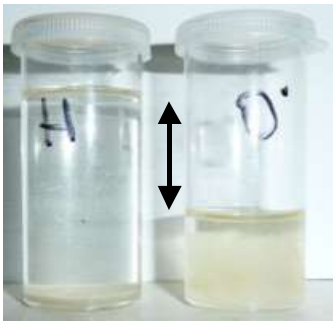

**Figure 4.** There is a swelling of starch in water with a different electric potential. Starch does not swell in water with the potential of –250 (left) and swells in water with the potential of +250 mV (right)

Water with a positive potential has an abnormal penetration, due to which it can evaporate even from a closed plastic bottle: the arrow shows how much during the day the level of such water has decreased.

Water with negative potential was obtained by bubbling uncharged water with hydrogen gas (left); water with positive potential was obtained by bubbling uncharged water with gaseous oxygen (right).

Both water used had 20 – 22 °C [4, 5].

It was also found that an increase in the positive electric potential of water is accompanied by an increase in its surface tension, and an increase in the negative electric potential of water is accompanied by a decrease in its surface tension. Since this relationship is very distinct, it is easily visualized (Figure 5) [4,5].

The fact that the surface tension of water strongly depends on its electric potential, salt crystals convincingly demonstrate. So, the drying of salt solutions prepared in water with a positive electric potential (charge) is accompanied by the formation of cubic or rhombic crystals (Figure 6, left), and the drying of salt solutions prepared in water with a negative electric potential (charge) is accompanied by the

 DOI: https://doi.org/10.30564/jhp.v2i1.1608

formation of needle or tree crystals (Figure 6, right) [4, 5].

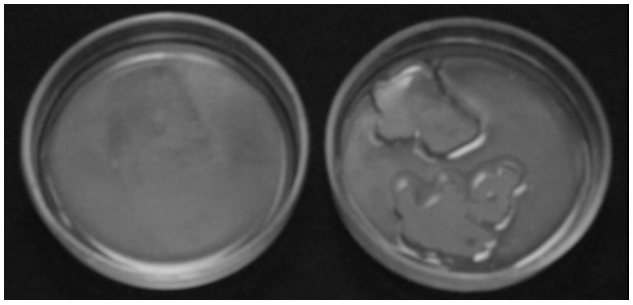

**Figure 5.** Left: 5 ml of water with a potential of –200 mV completely cover the bottom of the Petri dish. Right: 5 ml of water with a potential of +200 mV do not completely cover the bottom of a Petri dish

Due to the small surface tension, water with negative electric potential can spread throughout the bottom of the Petri dish; due to the large surface tension, water with a positive electric potential cannot spread throughout the bottom of the Petri dish [4, 5].

**Figure 6.** It is the crystals that formed after the drying of solutions of $KH_2PO_4$ prepared on the water with potentials of +250 mV (left) and –250 mV (right) [4,5]

It is especially important that the described regularities are subject to crystals formed during drying of aqueous solutions of sodium chloride (Figure 7), which is the main salt component of human biological liquids [5].

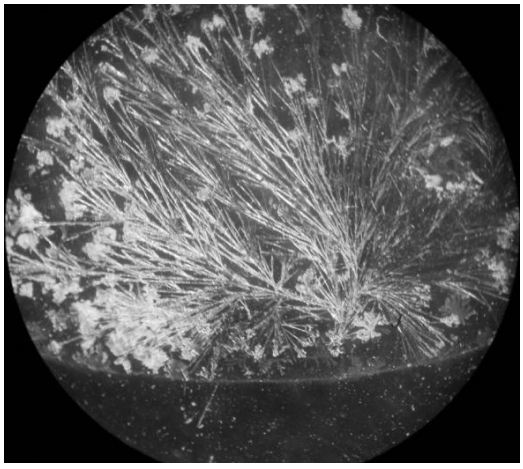

**Figure 7.** Crystals formed after drying a NaCl solution prepared in water with a potential of –200 mV [5]

Let us now try to evaluate all this knowledge in the medical aspect.

## 3. Discussion

### 3.1 The Relationship between the Electric Polarization of the Human Body and Its Hydration

It should be noted right away that the awareness of the described dependencies made it possible to assume that the properties of human biological fluids also substantially depend on their electric potential. So, this allowed us to explain the changes in the properties of biological fluids of women and the state of their reproductive organs during the menstrual cycle. Also, knowledge of the described dependencies made it possible to explain the polymorphism of crystals formed during the drying of biological fluids of women at different stages of the menstrual cycle [5].

The knowledge of these dependencies allowed us to assert that during the menstrual cycle there is a cyclic change in the electrical potential of biological fluids of women, in particular, that the beginning of the ovulation stage correlates with their negative electrization [5]. In addition, knowledge of the described dependencies allowed us to explain the nature of the phenomena that determine the weather dependence of the tone of blood vessels and human skin [6]. Thus, knowledge of the dependencies found to be very productive.

We now discuss the possible difference in the properties of biological fluids and the structures of a standing or sitting person, taking into account the electric polarization of his body. So, it can be assumed that the head of a standing person (together with the brain, vestibular apparatus, eyes, ears, etc.) is electrified positively, and its legs – negatively. As a result, the biological polymers located in the upper part of the human body are better hydrated than biopolymers located in its lower part. Since the hydration of biopolymers determines their stability, structure, and functional activity [7–10], it can be expected that the properties of biopolymers located in the upper and lower parts of a standing or sitting person will differ significantly.

This difference in the degree of hydration is especially important for DNA, whose properties strongly depend on the stage of hydration [11-13]. Suffice it to say that changing the degree of hydration of DNA molecules induces their A↔B transitions [11, 13] and, consequently, the activity of DNA and RNA polymerases, the intensity of peptide synthesis, cell proliferation, etc. [14].

In order to better understand the effect of electric polarization on human metabolism, it is useful to take into



DOI: https://doi.org/10.30564/jhp.v2i1.1608

account the results obtained by studying the effect of electrets on human cells, in particular on neuronal cells. In order to better understand the effect of electric polarization on human metabolism, it is useful to take into account the results obtained by studying the effect of medical electrets on human cells, in particular on neuronal cells. It is especially useful to take into account the fact that positively charged electrets initiate the proliferation of cells of the nervous system, and negatively charged electrets initiate the formation of dendrites by neurons [15] (compare with Figure 7).

Thus, standing on your head (Figure 8), yoga does not only change the usual view of the world.

**Figure 8.** This is Sirshasana or a headstand. When performing such an asana, negative charges accumulate in the person's head, and positive charges in his legs

There is no doubt that the tradition of doing a daily headstand is ancient. This allows us to conclude that the positive impact of this kind of depolarization on humans has been tested by time. In particular, it can be assumed that a daily headstand contributes to the formation of new dendritic processes in brain cells and is thus a way of counteracting dementia. There is also no doubt about the regenerative effect of the tradition of yogis on the brain to go to bed with the head on the equator, that is, in the direction of movement of negative charges. Thus, yogis have long used the distributive effect on the electric charges of both Lorentz forces $F_L$ and $F_L^*$ (Figure 1, see comments).

### 3.2. The Use of the Polarizing Action of the Considered Lorentz Forces in Manual Therapy

Let's also try to use the forces of Lorentz $F_L$ and $F_L^*$ for physiotherapeutic purposes, in particular, to increase the effectiveness of manual therapy. For this, we first take into account the electric polarization of human hands, namely, the negative electrization of the right hand and the positive electrization of the left hand (Figure 9).

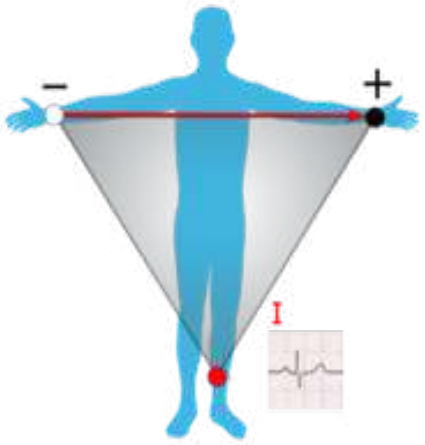


**Figure 9.** Human hands are polarized in such a way that the left hand has a predominantly positive charge, and the right one is predominantly negative [16]

In addition, it is necessary to take into account the location of the spinal nerves and their "areas of responsibility" (Figure 10).

This allows us to conclude that the Lorentz force $F_L$ will effectively direct the negative charges of the right hand (Figure 9) to the human spinal nerves (Figure 10) when it lies face down. Thus, we can stimulate the innervation of the corresponding organs and initiate the appearance of new interneuron contacts.

Obviously, directing negative charges of the right hand to the spinal nerves of a person can be no less effective if his face is oriented to the equator – in this case, the action of the Lorentz force $F_L^*$ will be used. Thus, in both cases we can get effects comparable to the effects of negatively charged electrets [15].

Clearly, also using the Lorentz force $F_L^*$, one can direct the positive charges of the left hand (Figure 9) onto the spinal nerves of a person whose face is oriented opposite to the equator. Thus, we can get an effect comparable to the effect of positively charged electrets [15].

 DOI: https://doi.org/10.30564/jhp.v2i1.1608

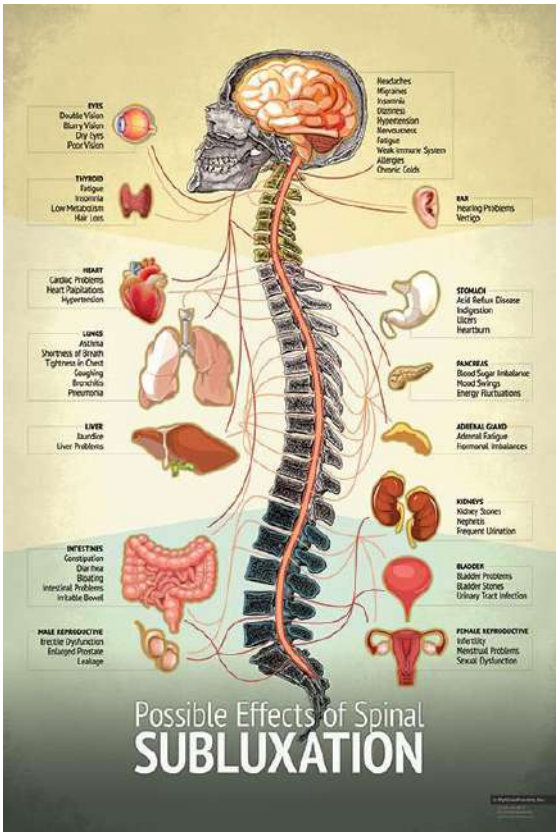


**Figure 10.** This shows the location of the head and vertebral nerves, as well as some of the organs that they innervate. Based on this, it is possible, in particular, to assess the consequences of injuries (curvature, dislocation, contusion, etc.) of different parts of the spine

Thus, taking into account the electric polarity of human hands (Figure 9) and using the Lorentz forces $F_L$ and $F_L^*$, the therapeutic effects of medical electrets [15] and pulsed electromagnetic fields [17, 18] can be reproduced.

## 4. Conclusion

The study of electromagnetic forces acting on the Earth must be continued. This need is motivated by an increase in the number of sources of electromagnetic radiation, both natural and artificial origin. It is also necessary to study the various effects of electromagnetic forces on humans, in particular their electrifying effect. It is hoped that the information obtained in these studies will help to better understand the causes of personal sensitivity to electromagnetic influences, both natural and artificial. One can also hope that this information will provide methods that can reduce personal sensitivity to electromagnetic influences. At the same time, the information obtained can be successfully used in preventive medicine, as well as give an understanding of the conditions for comfortable work and rest. Of course, the same information can be effectively used in manual and physiotherapy.

## References

[1] Pivovarenko Y. A Charge Distribution in the Earth's Atmosphere. American Journal of Physics and Applications, 2015, 3(3): 67-68.

[2] Pivovarenko Y. The Nature of the Celestial Elves, Sprites and Jets. Discovery Nature, 2018, 12: 1-4.

[3] Feynman R., Leighton R, Sands M. FLP, 2. Boston, Massachusetts: Addison Wesley Publishing Company. Sixth printing, 1977: 566.

[4] Pivovarenko Y. ±Water: Demonstration of Water Properties, Depending on its Electrical Potential. World Journal of Applied Physics, 2018, 3(1): 13-18.

[5] Pivovarenko Y. The Electric Potential of the Female Body Liquids and the Effectiveness of Cloning. Research and Reviews on Healthcare: Open Access Journal, Lupine Publishers, LLC, 2018, 1(2): 22-26.

[6] Pivovarenko Y. The Value of Gaseous Hydrogen Generated by the Intestinal Microflora of Human. Chapter 07 in: Top 10 Contributions on Biomedical Sciences: 2nd Edition, 2018: 2-15.

[7] Levitt M., Park B.H. Water: now you see it, now you don't. Structure, 1993, 1: 223-226.

[8] Eisenberg H. Protein and nucleic acid hydration and cosolvent interactions: establishment of reliable baseline values at high cosolvent concentrations, Biophys. Chem., 1994, 53: 57-68.

[9] Gerstein M., Chothia C. Packing at the protein-water interface. Proc. Natl. Acad. Sci. USA, 1996, 93: 10167-10172.

[10] Durchschlag H., Zipper P. Comparative investigations of biopolymer hydration by physicochemical and modeling techniques. Biophysical Chemistry, 2001, 93: 141-157.

[11] Kursar T., Holzwarth G. Backbone Conformational Change in the A-B Transition of Deoxyribonucleic Acid. Biochemistry, 1976, 15(15): 3352- 3357.

[12] Leal C., Wadso L., Olofsson G., Miguel M., Wennerstro H. The Hydration of a DNA-Amphiphile Complex. J. Phys. Chem. B, 2004, 108: 3044-3050.

[13] Pivovarenko Y. The Electric Potential of the Tissue Fluids of Living Organisms as a Possible Epigenetic Factor. Chemical and Biomolecular Engineering, 2017, 2(3): 159-164.

[14] Saenger W. Principles of Nucleic Acid Structure. New York - Berlin - Heidelberg - Tokyo: Springer-Verlag, 1984: 556.

[15] Alexandrova O.I., Aleksandrova S.A., Khomutov V.P., Morgunov M.S., Blinova M.I. Viability of cells of various types cultured on the surface of a medical electret. Journal of Technical Physics, 2018, 88(9):



DOI: https://doi.org/10.30564/jhp.v2i1.1608

1348-1354. (In Russian)

[16] Lilly L. S. ed. Pathophysiology of Heart Disease: A Collaborative Project of Medical Students and Faculty (sixth ed.). USA, Philadelphia: Lippincott Williams & Wilkins, 2016: 478 p.

[17] Goodwin T. J. Physiological and Molecular Genetic Effects of Time-Varying Electromagnetic Fields on Human Neuronal Cells; Technical Report of NASA. Lyndon B. Johnson Space Center Houston, Texas, 2003: 30.

[18] Pivovarenko Y. Arborization of Aqueous Chlorides in Pulsed Electromagnetic Fields as a Justification of Their Ability to Initiate the Formation of New Neuronal Dendrites. International Journal of Neurologic Physical Therapy, 2019, 5(1): 21-24.

 DOI: https://doi.org/10.30564/jhp.v2i1.1608

BILINGUAL PUBLISHING CO.
Pioneer of Global Academics Since 1984

**Journal of Human Physiology**
https://ojs.bilpublishing.com/index.php/jhp

ARTICLE

# Acute Effect of Kapalbhati Yoga on Cardiac Autonomic Control Using Heart Rate Variability Analysis in Healthy Male Individuals

**Rajeev Gupta***

Department of Neurology, All India Institute of Medical Sciences, New Delhi, India

ARTICLE INFO

*Article history*
Received: 17 March 2020
Accepted: 17 April 2020
Published Online: 31 May 2020

*Keywords:*
Autonomic nervous system
Sympatho-vagal balance
Cardiac autonomic control
Electrocardiography
Myocardial Ischemia

ABSTRACT

Kapalbhati is well known for improving cardiovascular health. But there are some reports of heart attacks while practising kapalbhati. We hypothesize that ill-effect of kapalbhati could be because of autonomic dysfunction to heart. In the present study, we aim to understand the acute effect of kapalbhati yoga on heart rate dynamics using heart rate variability (HRV) analysis. Resting heart rate (HR) varies widely in different individuals and during various physiological stresses, particularly, exercise it can go up to three-fold. These changes in heart rate are known as heart rate variability (HRV). Variability in heart rate reflects the control of autonomic system on the heart and which can be determined during brief periods of electrocardiographic (ECG) monitoring. HRV measures the effect of any physical exercise on the heart rate using time- and frequency-domain methods. Frequency-domain method involves power spectral analyses of the beat-to-beat intervals (R-R intervals) variability data. When total power vs. frequency, fast fourier transform analysis of R-R intervals data is done, it shows three well-defined peaks/rhythms in every individual, which contain different physiological information. Thus, the total spectral power of R-R intervals data can be divided into three components or bands viz., the very low frequency (VLF) band, the low-frequency (LF) band and the high frequency (HF) band. VLF represent very long time-period physiological phenomenon like thermoregulation, circadian rhythms etc. and thus are not seen in short-term recordings like in this work. LF band power represents long period physiological rhythms in the frequency range of 0.05-0.15 Hz and LF band power increases as a consequence of sympathetic activation. HF band represent physiological rhythms in the frequency range of 0.15-0.5 Hz and they are synchronous with the respiration rate, and arise due to the intrathoracic pressure changes and mechanical vibrations caused by the breathing activity. In this work, twenty healthy male volunteers were trained in kapalbhati yoga and their ECG waveforms (2 min.) were obtained while doing kapalbhati (breathing at 1 Hz frequency for 2 min.) and were compared with the baseline (just 2 min. before the start) and post-*kapalbhati* (immediately 2 min. after completing the practice) HRV data. Our results showed a significant decrease in the time-domain measures i.e., NN50, pNN50 and the mean heart rate interval during-*kapalbhati* when compared statistically to the respective before practice or "pre"-kapalbhati ($p < 0.05$, student's paired t-test) values. Frequency-domain indices showed that during-kapalbhati there is a significant increase (~48%) in the LF band power which suggests sympathetic activation and a significant increase (~88%) in the low frequency to the high frequency power ratio (LF/HF ratio) which indicates sympathetic system predominance. A significant decrease (~63%) in the HF component was also noted during-*kapalbhati* as compared to the "pre-*kapalbhati*" values which shows decrease in parasympathetic tone. Thus, these results suggest that during-*kapalbhati* there is drastic increase of sympathetic tone whereas parasympathetic activity is reduced. We propose these changes in autonomic system control on heart are responsible for the myocardial ischemic attacks induced during kapalbhati in some individuals.



DOI: https://doi.org/10.30564/jhp.v2i1.1755

## 1. Introduction

Yoga is a union of body and the mind. Kapalbhati is a fast (high frequency) yogic breathing technique which involves short, strong and rapid forceful exhalations at the rate of 1 to 2 Hz and inhalation is automatic such that mind is directed to the flow of breath [1]. Regularly practicing kapalbhati improves cardiac and mental health which makes it a very popular technique [2-6]. Despite reported health benefits, there are case reports available, suggesting people had undergone myocardial ischemic attacks while performing kapalbhati [7,8]. In a study, a decrease in cardiac vagal tone during kapālabhāti was reported due to decreased sensitivity of arterial baroreflex and increase in systolic blood pressure and low frequency blood pressure oscillations [7]. In another case, spontaneous pneumo-thorax caused by kapālabhāti was reported [8]. As Kapalbhati is a very popular technique for improvising cardiovascular health and practised widely by people of all age groups (especially by old age), it is very important to determine its deleterious effect on cardiovascular function if any. Intense exercisers say that watching heart rate variability (HRV) during any physical excercise can give an edge and a boost in workout performance. When HRV returns to normal after exercise, this shows that the person has fully recovered from workout, which tells it's safe to exercise again and helps avoid overtraining. Thus, HRV is a good indicator to study any ill-effects of kapalbhati on cardiac function. In this work, we aim to study the acute effect of kapalbhati practice on cardiac autonomic tone using HRV analyses while kapalbhati is done, so that maximum health benefits can be explored by every individual without deleterious side-effects. HRV analysis involves assessment of successive heart beat periods (between successive ECG wave R-R peak-to-peak intervals) using time- and frequency-domain methods. Time-domain measures involve measurement of different variables on the beat-to-beat intervals data. Frequency-domain indices involve decomposing R-R intervals into power-frequency spectrum which divides R-R intervals into three spectral components/bands viz., very low frequency band; the high frequency (HF) (0.15-0.5 Hz) and the low frequency (LF) (0.05-0.15 Hz) band [9-11]. Although there are several controversies in the scientific community regarding using LF power to indicate sympathetic activation but still till date it is used for the same [12]. On the other hand, HF band power and time-domain measures mainly reflect parasympathetic tone. Few reports are available on the effect of kapalbhati on cardiac function using HRV analyses. Raghuraj *et al* studied the effect of kapalbhati on autonomic function using HRV analyses before and after the practice using twelve male volunteers. They found significant increase in the LF power and LF/HF power ratio and significant decrease in the HF power immediately after doing 1 min. kapalbhati (at 2 Hz) when compared to the immediately before or "pre"-kapalbhati values [13]. Telles *et al* studied the effect of kapalbhati on HRV using thirty-eight male volunteers and found a significant decrease in NN50, pNN50 and the mean RR intervals while and after doing three rounds of 5 min. kapalbhati at 2 Hz but they found no change in any of the frequency domain measures during and after kapalbhati [14]. The above reports suffer from several limitations. First, they measure the effect of kapalbhati practice before and after the practice not during kapalbhati. Second, reproducibility of their results because of few reports is highly questionable. Thus, the present study is designed to study the acute effect of kapalbhati yoga on heart rate using HRV analyses during its practice.

## 2. Materials and Methods

### 2.1 Subjects

Twenty healthy young untrained male volunteers (age range, 18-35 years) not suffering from any known cardiovascular or neurological disorder and are not taking any medication were selected for the study. They were trained by a yoga instructor for one week in 1 Hz kapalbhati using a stop watch before the actual experiment. We made sure that participants did not have tea, coffee or meals at least two hours before participating in the study.

### 2.2 Recording

Volunteers were asked to sit in padmasana with back erect, rigid and wrist over the knee. Two ECG chest leads were placed below the collar bone on the left and right side and third one on the left fifth intercostal space at the mid axillary line ($V_6$). Baseline ECG waves

**Corresponding Author:*
*Rajeev Gupta,*
*Department of Neurology, All India Institute of Medical Sciences, New Delhi, India;*
*Email: scifigupta@gmail.com*

 DOI: https://doi.org/10.30564/jhp.v2i1.1755

(pre-*kapalbhati* data) were recorded with normal breathing for 2 minutes in the steady state immediately before starting kapalbhati and stored at 250 Hz frequency using portable Alice PDX polysomnography instrument (Philips-Respironics, USA). Then, volunteers were asked to do kapalbhati for 2 minutes (60 expirations in 1 min.) i.e. at 1 Hz frequency and ECG waves were continuously recorded (During-*Kapalbhati* data). After that, subjects took normal breaths, keeping the posture erect and ECG was recorded continuously for another 2 minutes (post-*kapalbhati* data). Thus, it took a total of six minutes to complete the entire recording from one individual and recording is done only once per volunteer. All the experiments were performed between 10:00 to 12:00 a.m. every time. ECG amplitude data (in μV) of pre-, during- and post-*kapalbhati* was separately exported into separate ASCII files and analyzed. The recording during-*kapalbhati* was collected only for 2 minutes because our aim in this study is to measure HRV during-*kapalbhati*, which can only be done continuously for two minutes by the recruited individuals.

### 2.3 Analysis

HRV was analysed in the pre-, during- and post-*kapalbhati* ECG data. ECG waveform was reconstructed from the amplitude data using the Kubios 2.2 HRV software (Biomedical Signal Analysis Group, Department of physics, University of Kuopio, Finland) and analysis was done using the time- and frequency-domain HRV methods. Power spectrum was separated into low frequency (LF, 0.04 – 0.15 Hz) and high frequency (HF, 0.15 – 0.4 Hz) components and a ratio of the LF and HF expressed in normalized units (n.u) (LF n.u./HF n.u.) was calculated. The following time-domain measures were analyzed: (i) mean R-R interval (the mean of the intervals between adjacent QRS complexes), (ii) mean heart rate, (iii) NN50 (the number of interval differences of successive NN intervals greater than 50 milliseconds), (iv) pNN50 (the proportion derived by dividing NN50 by the total number of RR intervals) in the 2 min. recordings. All the changes between the pre-, during-, post-*kapalbhati* were compared using paired student t-test. A p-value < 0.05 was considered statistically significant. All statistical analysis was done using Graph pad prism software.

## 3. Results

Table 1 shows comparison of different HRV variables pre-, during- and post-*kapalbhati*.

**Table 1.** Comparison of heart rate variability pre-, during- and post-*kapalbhati* in healthy male individuals

| Variables | Pre-Kapalbhati (n=20) | During-Kapalbhati (n=20) | Post-Kapalbhati (n=20) |
|---|---|---|---|
| Heart rate (bpm) | 73.59 ± 1.89 | 92.63 ± 1.62 | 77.00 ± 1.64 |
| LF (nu) | 38.31 ± 4.58 | 73.50 ± 3.67 | 55.74 ± 4.31 |
| HF (nu) | 53.49 ± 4.59 | 19.57 ± 2.80 | 39.15 ± 4.02 |
| LF/HF (nu) | 0.69 ± 0.19 | 5.88 ± 1.61 | 1.78 ± 0.88 |
| Mean RR (seconds) | 0.87 ± 0.02 | 0.66 ± 0.02 | 0.85 ± 0.03 |
| $NN_{50}$ (count) | 31.5 ± 9.46 | 7.3 ± 4.11 | 21.7 ± 6.09 |
| $pNN_{50}$ | 33.67 ± 5.23 | 5.40 ± 2.59 | 30.36 ± 4.43 |

***Notes:*** All values are expressed as Mean +/- Standard Error. bpm - beats per minute; LF – Low Frequency; nu – normalized units; HF – High Frequency.

### 3.1 Time-domain Analysis

Table 1 shows mean ± Standard Error values (n = 20) of mean heart rate; pre, during and post-*kapalbhati*. During-*kapalbhati*, mean heart rate increased by 19 beats per minute from pre-*kapalbhati* values. Pre-*kapalbhati* mean heart rate data was statistically compared with during-*kapalbhati* mean heart rate data using paired student t-test. The two-tailed p-value came out to be 0.0006 which is < 0.05 which suggests that the change in mean heart rate is significant. Mean R-R interval, NN50 and pNN50 were also computed. During-*kapalbhati*, a significant decrease in mean R-R interval, NN50 and pNN50 was noted as compared to "pre-" values. The decrease in mean RR intervals was extremely significant (p-value 0.0001<0.05). The decrease in NN50 was also found to be very significant (p-value 0.0046 <0.05).

### 3.2 Frequency-domain Analysis

Table 1 shows mean ± Standard Error values (n = 20) of normalized LF power, pre, during and post-*kapalbhati*. It is noted that there is an increase in LF power during-*kapalbhati* when compared with the pre-values. When the difference was checked for statistical significance between pre- and during-*kapalbhati* LF power, the p-value was less than 0.0001 which is < 0.05 suggesting extremely significant difference between the two LF power. Similarly, a reduction in HF values was observed during-*kapalbhati* as compared to the pre-values. When the difference was checked for statistical significance between pre- and during-*kapalbhati* HF power, the p-value was less than 0.0001 which suggests extremely significant difference. The increase in LF and reduction in HF band power lead to a significant increase in the LF/HF



DOI: https://doi.org/10.30564/jhp.v2i1.1755

ratio during-*kapalbhati* as compared to the baseline.

## 4. Discussion

The aim of the present study is to understand the control of autonomic nervous system on heart during kapalbhati practice. Heart rate variability (HRV) analysis is used to study the aim. A HRV analysis involves calculations on beat-to-beat intervals (R-R intervals) data using time- and frequency-domain methods. Time-domain methods (i.e., the mean R-R interval, NN50 and pNN50) which mainly reflect parasympathetic or vagal tone were calculated pre, during and post-*kapalbhati* practice. We observed a statistically significant decrease in the mean heart rate, mean RR interval, mean NN50 and mean pNN50 values during-*kapalbhati* as compared to the "pre-values" which suggest that during-*kapalbhati* there is decrease of parasympathetic autonomic tone. Frequency-domain measures (LF power, HF power and LF/HF nu power ratio) were also calculated. LF/HF nu ratio which is an indicator of balance between sympathetic and parasympathetic activity (also known as sympatho-vagal balance) is also calculated. Increase in LF power indicates sympathetic activation although its physiological interpretation is still controversial; it is thought both sympathetic and parasympathetic contributions can be involved in LF power activity. On the other hand, HF power mainly reflects parasympathetic activity. Under control or resting conditions, parasympathetic activity is the main activity which controls the heart rate and sympathetic activity is very low. In this study, we observed a statistically significant increase in the LF power during-*kapalbhati* as compared to the "pre-values" which signifies sympathetic system activation by the act of kapalbhati. Also, LF/HF power ratio increased drastically during-*kapalbhati* from "pre-values" and remained elevated post-*kapalbhati*, this shows that kapalbhati practice leads to sustained sympathetic activation and sympatho-vagal balance is disturbed which can be detrimental to heart if maintained for prolonged intervals. Furthermore, a decrease in the HF component was observed both during and immediately post-*kapalbhati* which suggests sustained inhibition or reduction of the parasympathetic activity to the heart. We propose over-activation of the sympathetic nervous system and withdrawl/inhibition of the parasympathetic activity is responsible for the deleterious cardiovascular effects reported due to kapalbhati practice reported in some individuals [7,8]. Heart rate is controlled by various neuronal and hormonal factors. Heart rate is determined by the rate of depolarization of the cardiac pacemaker. Pacemaker tissue is found in the sinoatrial (SA) node, the atrioventricular (AV) node, and the Purkinje tissue of the heart. But, SA node is considered the main pacemaker as it depolarizes faster than other pacemaker tissues and mainly controls the heart rate. SA node cells receive neuronal input from both sympathetic and parasympathetic (vagal) nerve fibers. Parasympathetic input to SA node decreases heart rate whereas sympathetic input increases it. During exercise, sympathetic activation leads to the release of catecholamines mainly epinephrine and norepinephrine from the post-ganglionic sympathetic nerve fibers innervating the heart which act on the SA node cells adrenergic receptors causing cells depolarization leading to increase of the heart rate. Also, norepinephrine is synthesized in the adrenal medulla which upon stimulation during physical, mental or emotional stress is released in the blood. Norepinephrine while in the blood circulation acts on all the body tissues including the heart. Norepinephrine is also released in the blood as a result of spillover (known as norepinephrine spillover) from the post-ganglionic sympathetic nerve fibers. Thus, norepinephrine exerts its sympathetic effect by acting both as a neurotransmitter and a hormone, thereby, increasing the heart rate during excercise. We think during kapalbhati yoga practice physical stress is induced which release norepinephrine both via the sympathetic nervous system to the heart and also from the adrenal medulla in the blood. In the heart, it acts on sinoatrial node (SA node) increasing the heart rate and in the blood stream it cause constriction of most of the blood vessels leading to increase of blood pressure. In the same way, parasympathetic activity to the heart is primarily mediated by post-ganglionic vagal nerve endings. Vagal nerve endings upon stimulation release acetylcholine neurotransmitter which acts on SA node slowing its conduction thus actively modulating vagal tone and slowing/relaxing the heart rate. This is to note that similar to our results an increase in the LF/HF power ratio and a decrease in the HF power has been reported previously immediately before myocardial ischemic attacks in coronary artery disease patients [15].

The exact mechanism by which doing kapalbhati influences autonomic tone to heart remains a question. It has been proposed earlier that at relatively high respiration rate of 60 $min.^{-1}$, hyperventilation might lead to changes in blood gases or pH which might stimulate central respiratory control. The respiratory and cardiovascular centers are closely associated in the brain stem. Thus, it is very likely that voluntarily changes in respiratory rate like while doing kapalbhati can bring about changes in cardiovascular parameters. As we have observed during kapalbhati there is sympathetic activation which

 DOI: https://doi.org/10.30564/jhp.v2i1.1755 

can increase heart rate and systolic blood pressure thus if it is performed beyond a certain time and at greater frequency can be detrimental to heart. In medical practice, hyperventilation (high frequency breathing such as kapalbhati) is used as a provocation method for the induction of seizures in suspected epileptic individuals. Thus, hyperventilation procedures are good for health only when performed with proper guidelines and the effect is highly individual-specific. All these factors justify our effort to measure HRV during kapalbhati, thereby helping in standardization of parameters (frequency, duration) for which if kapalbhati is done would provide maximum benefit to a particular individual. There are several limitations to our study like it has been done on healthy, young male individuals although kapalbhati is very popular among obese individuals and also amongst people of all age groups. Thus, this study needs to be repeated in individuals with cardiovascular disorders, obese people and people of all age groups where the results can be very different or even opposite. Furthermore, in this study we have used only HRV analyses as an indicator to study the effect of kapalbhati on cardiovascular function but there are several other cardiovascular parameters as well like baroreflex sensitivity, continuous blood pressure monitoring which should also be studied during kapalbhati practice to understand the complex mechanisms of the cardiac autonomic control from the research perspective.

## Conflict of Interest

There is no academic or financial conflict of interest.

## Funding Source

Dr. Rajeev Gupta is thankful to the Department of Biotechnology, Government of India for providing the Research Associate fellowship for carrying out this work.

## References

[1] Behanan, K. T.. Yoga, a scientific evaluation, 1960.

[2] Dinesh, T., Gaur, G. S., Sharma, V. K., Madanmohan, T., Harichandra Kumar, K. T., & Grrishma, B.. Effect of 6 weeks of Kapalabhati Pranayama training on peak expiratory flow rate in young, healthy, volunteers. Sch Acad J Biosci, 2013, 1(4): 111-114.

[3] Karambelkar, P. V., Deshapande, R. R., & Bhole, M. V.. Some respiratory studies in respect of' kapalabhati' and voluntary hyperventilation.Yoga Mimamsa, 1982, 21: 54-58.

[4] Kekan, D. R.. Effect of Kapalbhati Pranayama on Body Mass Index and Abdominal Skinfold Thickness. Indian Medical Gazette, Alternative Medicine, 2013, 431: 421-425.

[5] Kekan, D., Kashalikar, S.. Effect of Kapalbhati pranayama on waist and hip circumference. J Evol Med Dental Sci, 2013, 2(11): 1695-1699.

[6] Tyagi, A., Cohen, M.. Oxygen consumption changes with yoga practices: a systematic review. Journal of Evidence-Based Complementary & Alternative Medicine, 2013, 18(4): 290-308.

[7] Stancak Jr, A., Kuna, M., Vishnudevananda, S., Dostalek, C.. Kapalabhati--yogic cleansing exercise. I. Cardiovascular and respiratory changes. Homeostasis in health and disease: international journal devoted to integrative brain functions and homeostatic systems, 1990, 33(3): 126-134.

[8] Johnson, D. B., Tierney, M. J., Sadighi, P. J.. Kapalabhati Pranayama: Breath of Fire or Cause of Pneumothorax?: A Case Report. Chest, 2004, 125(5): 1951-1952.

[9] Stauss, H. M.. Heart rate variability. Am. J. Physiol. Regul. Integr. Comp. Physiol. 2003, 285(5): R927-R931.

[10] Nunan, D., Sandercock, G. R., & Brodie, D. A.. A quantitative systematic review of normal values for short-term heart rate variability in healthy adults. Pacing Clin. Electrophysiol. 2010, 33(11): 1407-1417.

[11]Akselrod, S., Gordon, D., Ubel, F. A., Shannon, D. C., Berger, A. C., Cohen, R. J.. Power spectrum analysis of heart rate fluctuation: a quantitative probe of beat-to-beat cardiovascular control. Science, 1981, 213(4504): 220-222.

[12] Goldstein, D. S., Bentho, O., Park, M. Y., & Sharabi, Y.. Low-frequency power of heart rate variability is not a measure of cardiac sympathetic tone but may be a measure of modulation of cardiac autonomic outflows by baroreflexes. Exp. Physiol. 2011, 96(12): 1255-1261.

[13] Raghuraj, P., Ramakrishnan, A. G., Nagendra, H. R., Telles, S.. Effect of two selected yogic breathing techniques on heart rate variability. Indian J. Physiol. Pharmacol. 1998, 42: 467-472.

[14] Telles, S., Singh, N., Balkrishna, A.. Heart rate variability changes during high frequency yoga breathing and breath awareness. Biopsychosoc. Med. 2011, 5(1): 4.

[15] Goseki, Y., Matsubara, T., Takahashi, N., Takeuchi, T., Ibukiyama, C.. Heart rate variability before the occurrence of silent myocardial ischemia during ambulatory monitoring. Am. J. Cardiol. 1994, 73(12): 845-849.



DOI: https://doi.org/10.30564/jhp.v2i1.1755

## Supplementary Data

**Table 2.** Mean ± Standard Error values (n = 20) of heart rate (HR) pre-, during- and post-*kapalbhati* yoga

| S. No. | Heart Rate pre-Kapalbhati | Heart Rate during-Kapalbhati | Heart Rate post-Kapalbhati |
|---|---|---|---|
| 1. | 62.86 | 90.32 | 70.58 |
| 2. | 64.28 | 89.26 | 60.54 |
| 3. | 82.91 | 92.09 | 81.91 |
| 4. | 76.91 | 98.02 | 81.68 |
| 5. | 86.26 | 91.23 | 75.90 |
| 6. | 60.20 | 83.67 | 75.64 |
| 7. | 64.48 | 76.69 | 61.50 |
| 8. | 89.45 | 91.11 | 91.11 |
| 9. | 64.95 | 90.29 | 82.77 |
| 10. | 80.15 | 110.14 | 80.81 |
| 11. | 73.54 | 100.72 | 76.38 |
| 12. | 74.91 | 96.02 | 79.68 |
| 13. | 72.42 | 89.32 | 75.63 |
| 14. | 75.95 | 101.29 | 80.67 |
| 15. | 80.91 | 99.91 | 79.68 |
| 16. | 70.26 | 89.02 | 75.28 |
| 17. | 77.50 | 97.12 | 83.32 |
| 18. | 79.12 | 90.26 | 83.15 |
| 19. | 69.52 | 85.62 | 75.29 |
| 20. | 65.71 | 90.51 | 68.52 |
| | 73.59 ± 1.84 | 92.63 ± 1.62 | 77.00 ± 1.64 |

**Table 3.** Mean ± Standard error values (n = 20) of ECG RR intervals (in seconds) pre-, during- and post-*kapalbhati* yoga

| S. No. | RR intervals (in seconds) Pre-*kapalbhati* | During-*kapalbhati* | Post-*kapalbhati* |
|---|---|---|---|
| 1. | 0.95 | 0.63 | 0.87 |
| 2. | 0.93 | 0.67 | 1 |
| 3. | 0.72 | 0.65 | 0.74 |
| 4. | 0.78 | 0.61 | 0.73 |
| 5. | 0.74 | 0.67 | 0.8 |
| 6. | 1 | 0.75 | 0.84 |
| 7. | 0.94 | 0.78 | 0.97 |
| 8. | 1.16 | 0.91 | 1.21 |
| 9. | 0.93 | 0.66 | 0.87 |
| 10. | 0.75 | 0.5 | 0.77 |
| 11. | 0.90 | 0.60 | 0.82 |
| 12. | 0.88 | 0.62 | 0.96 |
| 13. | 0.72 | 0.64 | 0.74 |
| 14. | 0.75 | 0.59 | 0.68 |
| 15. | 0.95 | 0.75 | 0.79 |
| 16. | 0.93 | 0.77 | 0.96 |
| 17. | 0.92 | 0.65 | 0.86 |
| 18. | 0.74 | 0.50 | 0.76 |
| 19. | 0.86 | 0.67 | 0.85 |
| 20. | 0.89 | 0.58 | 0.82 |
| | 0.87 ± 0.02 | 0.66 ± 0.02 | 0.85 ± 0.02 |

**Table 4.** Mean ± Standard Error values (n = 20) of pNN50 (%) values pre-, during- and post-*kapalbhati* yoga

| S. No. | pNN50 (%) Pre-*kapalbhati* values | During-*kapalbhati* | Post-*kapalbhati* |
|---|---|---|---|
| 1. | 44.3 | 0 | 36.9 |
| 2. | 30.6 | 0 | 31.0 |
| 3. | 6.2 | 0 | 11.4 |
| 4. | 2.7 | 0 | 2.5 |
| 5. | 82.3 | 45.9 | 76.1 |
| 6. | 84.5 | 13 | 75.4 |
| 7. | 29.2 | 25.7 | 14.3 |
| 8. | 1.1 | 0 | 1.1 |
| 9. | 63.9 | 4.5 | 46.2 |
| 10. | 20.5 | 0 | 30.3 |
| 11. | 40.5 | 0 | 31.5 |
| 12. | 35.3 | 0 | 34.5 |
| 13. | 11.2 | 0 | 15.4 |
| 14. | 5.7 | 0 | 7.5 |
| 15. | 32.53 | 0 | 30.53 |
| 16. | 38.21 | 0 | 30.89 |
| 17. | 40.82 | 10 | 35.59 |
| 18. | 36.53 | 8.91 | 32.52 |
| 19. | 37.45 | 0 | 33.95 |
| 20. | 29.9 | 0 | 29.8 |
| | 33.67 ± 5.23 | 5.40 ± 2.59 | 30.36 ± 4.43 |

**Table 5.** Mean ± Standard Error values (n = 20) of normalized power of low frequency (LF) component of heart rate variability (HRV) pre-, during- and post-*kapalbhati* yoga

| S. No. | LF pre-Kapalbhati | LF during-Kapalbhati | LF post-Kapalbhati |
|---|---|---|---|
| 1. | 17.9 | 88.3 | 63.2 |
| 2. | 70.3 | 94.5 | 90.6 |
| 3. | 36.6 | 48.1 | 35.8 |
| 4. | 18.3 | 79.7 | 74.7 |
| 5. | 34.2 | 90.6 | 40.3 |
| 6. | 48.3 | 86.1 | 22.6 |
| 7. | 31.1 | 72.6 | 59.8 |

 DOI: https://doi.org/10.30564/jhp.v2i1.1755

| | | | |
|---|---|---|---|
| **8.** | 39.4 | 89.1 | 48.5 |
| **9.** | 20.3 | 63 | 49.9 |
| **10.** | 40.7 | 81.3 | 46.1 |
| **11.** | 84.24 | 94.50 | 91.27 |
| **12.** | 14.00 | 38.51 | 27 |
| **13.** | 17.7 | 71.0 | 63.5 |
| **14.** | 59.26 | 89.39 | 85.48 |
| **15.** | 75.5 | 80.6 | 69.1 |
| **16.** | 44.2 | 66.4 | 55.01 |
| **17.** | 35.98 | 56.05 | 40.71 |
| **18.** | 37.87 | 54.49 | 51.21 |
| **19.** | 22.7 | 65.39 | 50.29 |
| **20.** | 17.7 | 60.5 | 49.9 |
| | 38.31 ± 4.58 | 73.50 ± 3.67 | 55.74 ± 4.31 |

**Table 6.** Mean ± Standard Error values (n = 20) of high frequency (HF) power component of heart rate variability pre-, during- and post-*kapalbhati* yoga

| S. No. | HF pre-Kapalbhati | HF during-Kapalbhati | HF post-*kapalbhati* |
|---|---|---|---|
| **1.** | 81.7 | 11.6 | 36.7 |
| **2.** | 29.7 | 5.5 | 9.4 |
| **3.** | 62.6 | 51.6 | 63.5 |
| **4.** | 81.4 | 20.2 | 25.2 |
| **5.** | 65.7 | 9.4 | 59.6 |
| **6.** | 51.6 | 13.9 | 77.3 |
| **7.** | 68.9 | 27.2 | 40.2 |
| **8.** | 60.5 | 10.9 | 51.5 |
| **9.** | 79.1 | 36.8 | 49.3 |
| **10.** | 59.1 | 18.6 | 53.9 |
| **11.** | 54.49 | 37.87 | 51.21 |
| **12.** | 56.05 | 35.98 | 40.71 |
| **13.** | 15.73 | 5.49 | 8.71 |
| **14.** | 62.6 | 5.16 | 43.5 |
| **15.** | 34.38 | 17.5 | 33.20 |
| **16.** | 35.94 | 16.8 | 22.2 |
| **17.** | 37.89 | 23.8 | 36.33 |
| **18.** | 25.00 | 16.41 | 15.23 |
| **19.** | 28.52 | 15.23 | 27.7 |
| **20.** | 81.7 | 11.6 | 36.7 |
| | 53.63 ± 4.59 | 19.57 ± 2.80 | 39.15 ± 4.02 |



DOI: https://doi.org/10.30564/jhp.v2i1.1755

BILINGUAL PUBLISHING CO.
Pioneer of Global Academics Since 1984

**Journal of Human Physiology**
https://ojs.bilpublishing.com/index.php/jhp

**ARTICLE**

# Milestones of the Modeling of Human Physiology

**Grygoryan R.D.***

Head of department "Human systems modeling", Cybernetics Center, Institute of software systems of National Academy of Sciences, Kiev, Ukraine

ARTICLE INFO

*Article history*
Received: 21 May 2020
Accepted: 6 July 2020
Published Online: 17 August 2020

*Keywords:*
Computer simulations
Visualization
Theoretical Analysis
Quantification
Multi-scale modeling
Physiome

ABSTRACT

An overview of about 70-year research efforts in area of mathematical modeling of human physiology is provided. The overview has two goals: (1) to recognize the main advantages and causes of disadvantages or disappointments; (2) to distinguish the most promising approach for creating future models. Until recently, efforts in the modeling of quantitative physiology were concentrated on the solving of three main types of tasks: (1) how to establish the input-output dynamic characteristics of a given isolated organ or isolated anatomical-functional system (AFS); (2) how to create a computer-based simulator of physiological complex systems (PCM) containing many organs and AFSs; and (3) how to create multi-scale models capable of simulating and explaining causalities in organs, AFSs, PCMs, and in the entire organism in terms that will allow using such models for simulating pathological scenarios (the "Physiome" project) too. The critical analysis of the modeling experience and recent physiological concepts convinced us that the platform provided by the paradigm of physiological super-systems (PPS) looks like the most promising platform for further modeling. PPS causally combines activities of specific intracellular mechanisms (self-tunable but of limited capacities) with their extracellular enhancers. The enhancement appears due to the increase of nutrients incomes toward cells affected because of low energy and inadequate chemical composition of cytoplasm. Every enhancer has its activator chemicals released by the affected cells. In fact, PPS, indicating causal relationships between cell-scale and upper-scales (in organs, AFSs, PCMs) physiological activities, is the single platform for future models. They must definitely describe when and how the bottom-to-up information flows do appear and how is the organism-scale adaptation activated against destructive trends in cells.

## 1. Introduction

In traditional human and animal physiology, mathematical methods for a long time were associated and mainly limited by statistics and regression analysis. However, since the middle of the XX Century, researchers who were initially educated in mathematics, physics, engineering or cybernetics, started to collaborate with biologists, widened the opportunities for analyzing life events and living systems. In particular, the adjoined researchers started to apply their professional knowledge and skills to assist in solving of certain actual problems in human physiology and medicine. Over time, one group of

**Corresponding Author:*
*Grygoryan R.D.,*
*Head of department "Human systems modeling", Cybernetics Center, Institute of software systems of National Academy of Sciences, 03083, 53, Admiral Ushakov str., Kiev, Ukraine;*
*Email: rgrygoryan@gmail.com*

 DOI: https://doi.org/10.30564/jhp.v2i1.1905

these assistants, who would not change their initial specialization, again switched-out to solving other problems not related to biology. Others, on the contrary, began to go so deep into biology, in particular, into physiology, that they began capable to clarify the existent biological concepts or even independently propose new concepts that in turn became the object of mathematical modeling and verifying. In parallel with the increased computers' power, and with the advent of new computing technologies for visualizing complex dynamic processes of life, these neo-physiologists became independent researchers capable of self-working on the creation of theoretical human physiology. Certainly, concepts concerning mathematical modeling of physiological events, objects, and systems also have been evolved. Currently, there is none human organ or anatomical-functional system (AFS) that's function is not formally described yet and investigated via computer simulations. Currently, publications on different aspects of these investigations exceed tens of thousands of articles, thousands of dissertations and monographs. The purpose of this article is not to review all of these publications: it is simply impossible. Readers interested in modeling methods and specifics of concrete models can independently find the necessary information in thematic reviews.

The goals of this short review article are (1) in outlining of the main milestones and approaches to modeling of human physiology, and (2) in highlighting the most promising ways for creating of models that could simulate the human integrative physiology on a basis of main rules evolutionary saved in a community of specialized cells.

## 2. A Brief Prehistory

The phenomenon of life was of interest not only to thinkers of antiquity and natural philosophers of the seventeenth and eighteenth centuries. In the 1870-s, Claude Bernard suggested the physical-chemical state of the internal liquid environment of the body to be almost constancy. These conjectures were strictly experimentally substantiated by Walter Cannon in 1929 when he formulated his concept of homeostasis [1]. Cannon's achievements intensified the common interest to life phenomenon. The mathematician Norbert Wiener, who collaborated with the physiologist Arturo Rosenblueth, and not only published a joint article with him on the processes of excitation in the heart muscle [2], but also, using Cannon's idea of the stabilizing role of negative feedback, he created a new science - cybernetics [3]. Some earlier, the famous physicist, one of the creators of quantum mechanics, Erwin Schrödinger published a book in which he tried to explain life from the point of view of a physicist [4]. Two questions he formulated concerned the basis of life - why biological molecules have to be permanently synthesized and why life needs enormously high energy expenditures. Namely, later by answering these questions, biochemists gave physiologists a new key for explaining the fundamental causalities between energy support and physiology. Now we realize that the emergence of special mechanisms (we call them physiological mechanisms), using the outside energy for the organization of cyclic transformations against natural gradients, had supported the physics and chemistry to origin the life phenomenon.

Although these scientists could not solve all the problems of biology, we should consider them as first modelers of the life phenomena: they have had looked at life from novel perspectives and their ideas and research methodology were further developed in unexpected directions.

## 3. A Brief History of Human Physiology Modeling Approaches

Cybernetics streamlined both design and the mathematical methods of calculation of stable technical dynamic systems. By the beginning of the 1950-s, the interest of cybernetics-mathematicians in the phenomenon of life received a methodologically new research impulse. Its initiator was Fred Grodins [5-6], for the first time tried to use the methodology of the control theory for a mathematical synthesis of human cardiovascular and respiratory quantitative physiology.

Looking at the history of physiological objects' and processes' modeling from a *bird's-eye view*, we can distinguish three main phases each with its characteristic signs.

At the initial stage, which started in the middle 50-s and continued up to the 90-s of the XX Century, modelers have seen their main role in formalizing accumulated empirical data. As the data basis mainly concerned physiology of isolated organs (heart, kidneys, liver, and pancreas) or anatomical-functional systems (AFS) like the cardiovascular system (CVS) and the respiratory system (RS), these systems were first formalized [5-15]. The formalization, spirited by the theories of the cybernetics and automatic systems, was aimed to present the functioning of both CVS and RS in cybernetic terms. Internal structural nuances and their likely consequences on the functionality of the original object had not been considered. The object had been looked like a "black-box" that within the given time interval $\tau$ (observation time) has its input variables $X(x_1(t)\ x_2(t),...,x_m(t)$ , output variables $Y(y_1(t)\ y_2(t),...,y_n(t)$ , and internal constants



DOI: https://doi.org/10.30564/jhp.v2i1.1905

$K(k_1, k_2, ..., k_p)$. In the frame of this formalization, the function $Z(K,t)$ calculated as:

$$Z(K,t) = \frac{Y(y_1(t), y_2(t), ..., y_n(t); k_1, k_2, ..., k_p)}{X(x_1(t), x_2(t), ..., x_m(t))} \quad (1)$$

is a transfer function of the modeled object. Quantitative investigations of $Z(K,t)$ for certain test-situations (for variations of $x_1(t)$, $x_2(t), ..., x_m(t)$; $y_1(t)$, $y_2(t), ..., y_n(t)$; and\or $k_1, k_2, ..., k_p$) allowed the modeler to obtain additional calculated data capable to give the physiologist-empiricist additional information about object's properties.

In the sixteenths and seventeenths of the XX Century, one of the main physiological problems was a problem how to determine the goal functions of physiological systems (CVS, RS, or others) [15-17]. Moreover, echoes of this problem have not calmed down to this day too [18-22]. In this regard CVS is an illustrative object to demonstrate the novelty of the cybernetic analysis.

CVS provides organism by two variables - cardiac output (CO) and arterial pressure (AP). According to cardiovascular mechanics, the mean arterial pressure (MAP) better characterizes the hemodynamic laws than AP which is displaying cyclic variations. Therefore, experts often operate with MAP than with AP.

Physiologists discussed whether CO or MAP is the main goal function of CVS. Most experts suggested the homeostasis of MAP, provided by baroreceptor reflexes, to be the main goal function of CVS [18-23]. However, empirical data illustrated many situations in which MAP-level is floating [24-27]. One of actual problem was associated with hypertension [27-30]. According to spread concept, the altered sensitivity of MAP's control mechanisms is an initiator of hypertension thus certain theoretical research was focused on searching of quantitative approaches capable of measuring or calculating this sensitivity.

The cybernetic concept considers CVS to be a dynamic closed system (CS) built of two open systems (OS): an uncontrolled object (UO) and a regulator systems (RS). In fact, OS is the heart and vasculature filled by a summary blood volume and possessing by certain constant parameters. The function of RS is to alter these constants depending on input information came from multiple sensors. In short time intervals, arterial baroreceptors were commonly suggested to be the main sensors thus the arterial baroreflex is considered to be the main RS of MAP [13,16,21,26]. In long-time intervals, RS includes all physiological mechanisms capable of altering the hemodynamics. As the latter also depends upon summary blood volume that is under influences of multiple mechanisms and factors not relating to CVS, this simplified presentation is not to be adequate. Nevertheless, using the cybernetic approach, investigators have had calculated the values of gains for both physiological and hypertensive conditions [14-16].

Another approach to clinical use of calculated gains (sensitivity coefficients) was focused on assessing of the baroreflex servo-mechanism via its ability to change UO's parameters. As the heart rate ($F(t)$) is the most easy measurable parameter, the approach can be illustrated using $F(t)$. It is necessary to have at least two pairs of values of both MAP and heart rate: one - in an initial state of organisms, the second - after application of a standard procedure causing alteration of $F(t)$. For our convenience, let us denote MAP as $P_A$. Then $\Delta P_A = P_{A2} - P_{A1}$ and $\Delta F = F_2 - F_1$. The sensitivity of the heart rate to changes of MAP is: $S_F = \Delta F / \Delta P$. It is not hard to see that this approach requires all other influences on MAP and heart rate to be constant. However, evolution has saved a lot of multi-scale mechanisms simultaneously altering both these variables. Thus within the interval $F_{\min} \le F(t) \le F_{\max}$ real $F(t)$ should be calculated as:

$$F(t) = \begin{cases} F_{\min}, & \sum_{j=1}^{n} \Delta F_i^-(t) > F_a - F_{\min} \\ F_a + \Delta F(T) + \sum_{i=1}^{m} \Delta F_i^+(t) - \sum_{j=1}^{n} \Delta F_i^-(t), & F_{\min} \le F(t) \le F_{\max} \\ F_{\max}, & \sum_{i=1}^{m} \Delta F_i^+(t) + F_a > F_{\max} \end{cases} \quad (2)$$

where $F_a$ is the heart rate under normal temperature ($T$), biochemical characteristics of blood, and biophysical characteristics of cells of sinus node, $\Delta F(T)$ is elevation of $F_a$ with temperature increasing, $\Delta F_i^+(t)$ are accelerating effects of $m$ mechanisms and $\Delta F_j^-(t)$ are deccelerating effects of $n$ mechanisms. Note, that each such mechanism has its power and developmental inertia.

There is a theoretical problem: is each mechanism, influencing HR, specific regulator of CVS or certain mechanisms do not have the necessary and sufficient signs to be considered as the regulator [31-33]. Indeed, the baroreceptor reflexes that have receptor organs, central neurons reacting to the receptor impulses, and *sympa-*


DOI: https://doi.org/10.30564/jhp.v2i1.1905

*thetic* and *vagus* nerves for controlling the activity of sinus node pacemakers, form a closed negative feedback nervous-reflector loop. However, a lot of blood chemicals influencing $\Delta F_i^+(t)$ or $\Delta F_i^-(t)$, have not their own feedback loops. By the way, this note also concerns multiple mechanisms generally activating or suppressing CVS via humoral channels [34]. The last factor is one of causes forcing physiologists to think that certain chemical agents like angiotensin, vasopressin, NO, CO2, SO2, despite having real pressor or depressor effects, are sooner sources for the appearing hypertension or hypotonic state than regulators of MAP-level. Real CVS is not an isolated system as it is assumed in the most models of hemodynamics. CVS interacts with multiple organs and AFS. In particular, total blood volume ($V_S(t)$), that is the main modifier of both central venous pressure and MAP, only in very little values of the observation time $\tau$ can be considered to be constant. Modifiers of $V_S(t)$ are acting via changing the liquid intakes from the digestive system ($q_w(t)$), of the diuresis ($q_d(t)$), expirations in lungs and skin. So, these effectors obviously do not belong to CVS.

$$\frac{dV_S}{dt} = q_w(t) - q_d(t) - q_{es}(t) - q_{ee}(t) - q_{cf}(t) + C_{be}(t) + C_{bl}(t) \tag{3}$$

where $q_{cf}(t)$ - trans-capillary flows, $q_{es}(t)$ - the evaporation with sweat, $q_{ee}(t)$ - expiratory fumes, $C_{be}(t)$ - blood salt concentrations, $C_{bl}(t)$ - concentrations of blood lipids.

In addition, MAP-level is also under influences of vessels' rigidity ($D(t)$) and unstressed volume ($U(t)$). However, neither $D(t)$ nor $U(t)$ can be directly measured: at organism-scale, they are still virtual characteristics. Nevertheless, they are important for modeling, especially in models with lumped parameters [7-10,12,36,37]. Formally,

$$D(t) = D0 + \sum_{i=1}^{m1} \Delta D_i(t); \quad U(t) = U0 - \sum_{i=1}^{m1} \Delta D_i(t), \tag{4}$$

where $m1$ is the number of vascular region, $D0, U0$ represent the initial values of $D(t)$ and $U(t)$.

The considerations above, formally presented in (2)-(4), are not the exclusive problems arose during use the model-based methods in clinics. As a rule, the function $Z(K,t)$ for an organ or even for a receptor is non-stationary and has a non-linear form. In case of a multi-fiber aortic nerve, the non-linear impulses-pressure curve looks like S-shaped [38,39]. For correct calculations of gains one needs to use complex calculus. The problem is facilitated by the fact that in the field of normal physiological quantities of $x_1(t)\ x_2(t),...,x_m(t)$; $y_1(t)\ y_2(t),...,y_n(t)$, and $k_1, k_2,..., k_p$, the non-linearity can be ignored and linear dependencies used. In the simplest case, when $m = 1$ instead of the dynamic $Z(K,t)$ one can use a simple output-input ratio.

Modelers try to approximate empirically obtained nonlinear curves by piecewise linear ones. Though such substitutions often are not incorrect they do not reveal and take into account one important internal cause of the nonlinearity. The matter is that the nonlinearity usually is an effect appearing when one sums a lot of discrete results that are reactions of individual components (receptors, common type cells) forming a population [34]. In fact, elements of such populations rarely are absolutely identical. Normally, they have slight structural differences (anisotropies). So, changes in the statistical distributions of elements possessing by the same anisotropy altering the graph form of $Z(K,t)$.As these ultra-structural alterations are the first signs of pathological trends, the substitution of the dynamic $Z(K,t)$ by the simple output-input ratio hides these early pathological trends.

In (1), we considered $k_1, k_2,..., k_p$ to be independent of time. However, this assumption is not correct forever. In some organs (for example, in heart) certain $k_1, k_2,..., k_p$ are constant in the models that describe static input-output relations and variables if the model does simulate the dynamic events within the cardiac cycle. Thus in the latter case, the myocardium elastance, valves' resistance are included with approximations of their dynamics [40-44].

The cybernetic concept and calculations of the simplified version of the gain also were used during the analysis of more complex RSs like the renal control of MAP through the renin-angiotensin-aldosterone loops (see review [45]). However, there is an additional reason to estimate such theoretic models as those that creating incorrect physiological understanding of negative feedbacks. The matter is that cybernetics use a virtual notion known as "set-point". The cybernetic mechanism stabilizing the value of $Y(y_1(t)\ y_2(t),...,y_n(t)$ despite certain alterations of $X(x_1(t)\ x_2(t),...,x_m(t)$ is based on a comparison of the current value of $Y(t)$



DOI: https://doi.org/10.30564/jhp.v2i1.1905

with a given value of $Y_0$ (namely, the "set-point"). A difference $\varepsilon = Y_0 - Y(t)$ is a signal that goes to the gears forming a correction influence added to the input variable $X(x_1(t)\ x_2(t),...,x_m(t)$ . However, real neuronal structures providing the "set-point" have not been identified yet. So, reasonable is to state that biological homeostatic mechanisms will have another organizing principle. The reciprocally acting physiological mechanisms are the most likely pretenders of this role [34].

Perhaps, the reasons and examples described above are sufficient to state that already at the first phase of modeling, in several models most organs, instead of the "black-box", were already transformed into a "gray-box" in which certain physiological rules were already clarified and formalized. Certainly, this widened model's usability. At the same time, a lot of difficulties and problems appeared. Some of them practically disappeared with the beginning of modeling's second phase that began due to appearing of powerful personal computers. A little by little, new mathematical methods and software, had provided modelers by high-speed computing and advanced visualization technologies. So, modelers became aware of creating models containing more and more detailed physiological nuances.

During1980-s and early 1990-s, traditional physiology has essentially modified due to penetrated methods of molecular analysis. Therefore, the advanced versions of models increasingly included descriptions of the molecular basis of certain physiological mechanisms. However, already in the middle of 1990-s, both modelers and potential users met new problems: (1) how to compare simulation results of different versions of the one and the same organ's model; (2) how to estimate model's correctness; (3) how long the necessity of new model creation will last yet, and at last (4) is it possible to unify human physiology models?

The "Physiome" long-term project opened the third phase of modeling and lasting currently was the general answer to these questions.

### *The "Physiome" Ideology and Research*

The "Physiome" project was inspired by the well-known "Genome" project. The International Union of Physiological Sciences (IUPS) launched the Physiome Project at the IUPS World Congress in St Petersburg in 1997 [46]. Apologists of a novel research ideology aimed to bring quantitative bioengineering approaches to the study of anatomy and physiology. Both physiologists and modelers assumed that the model of human physiology must incorporate all scientific data concerning genetic, molecular, intracellular, multicellular events, organs' functionalities, and at last integrative organism-scale physiology. It also was declared every model to be strongly verified using empirical data. In the frame of the project, to support it by powerful techniques and technologies, both creation of special software and data centers for an accumulation of both initial biological and simulation data was planned [http://www.iups.org/physiome-project/]. Financial support of the research and developments was organized in the frames of both international "Physiome" projects (http://www.iups.org/physiome-project/; http://physiomeproject.org/about/molecules-to-humankind; https://www.auckland.ac.nz/en/abi/our-research/research-groups-themes/physiome-project.html), and national "Physiome" projects (for example, [46-52]; (https://www.physiome.org/;https://www.physiome.org/Links/publications.html; https://www.researchgate.net/publication/312381514_HD_Physiology_Project-Japanese_efforts_to_promote_multilevel_integrative_systems_biology_and_physiome_research.

Over the past two decades, in the development of models, the ideology of the Physiome has become to be dominant. Many models of particular physiological mechanisms that function on different spatial scales have been created (for example, [53-57]. We cannot provide here for their detailed and comparative analysis. We only note that each such model is a huge work of its creators, which includes not only complex mathematics and simulation technologies, but also a rigorous justification of the assumptions forced to made and the limitations resulting from them. As a rule, most models were created with a direct or remote applied purpose. Some of them are relatively simple, while the model [56], designed to simulate the main effects of a combination of physiological and artificial mechanisms protecting humans against loss of vision or consciousness in conditions of extreme pilotage accelerations, contains more than 2000 differential equations. Since none of the Earth-based physical models is adequate, separate models (for example [57,58]) were developed to test existing hypotheses regarding the processes of the organism's adaptation to zero-gravity conditions.

To our opinion, at least in the aspect of the multi-scale physiology, the "Physiome" ideology met unsolvable problems. Some of them are discussed below.

## 4. Discussion

Physiology is a fundamental science that simultaneously is the theory of medicine. However, physiologists already released that their empirical research methods are too


DOI: https://doi.org/10.30564/jhp.v2i1.1905

primitive to reveal the real complexity of the human organism. Models developed since the middle 1950-s use origin mixed reactions of traditional physiologists. On the one side, they need methodological support. On the other side, mainly because the mathematics and research technologies used by modelers are not comprehensively understood by physiologists, the latter feeling certain distrust in these models and the results. This distrust is also based on the fact that models use a lot of quantitative data that cannot be measured. In contrast, orders (users) of applied models use simulators to have certain presentations, namely about these un-measured living characteristics. Perhaps, this is why applied physiologists ordered the most computer simulators. It seems this reason is right also for the practical medicine needed to improve diagnostic and treat technologies via the creation of personalized computer models of pathologies. Namely, the medical needs forced to support large research projects like the "Physiome" project. However, to our opinion, models of complex physiological mechanisms are still very far from this medical ideal. Nevertheless, modeling efforts have to be supported because there is none acceptable alternative to how to reveal these mechanisms.

Only a little part of the models has been analyzed above. The analysis din not concern models of functional systems like the system of thermoregulation and the immune system. Certainly, models of both these systems also exist (for example, [59-62]).

### *Concerning the Reasonable Complexity of the Model*

The model cannot take into account all the facts. If one creates a didactic model of certain organ or AFS, he/she are free to reduce the model until it simulates distinguish biological functionalities. Currently, a lot of depositaries accumulated free-usable didactic models of human physiology (https://caehealthcare.com/patient-simulation/pediasim/; https://physiology.kitware.com/; https://biogearsengine.com/; http://hummod.org/; http://simvascular.github.io/; https://simtk.org/projects/opensim/). Creators of other models always are searching for a reasonable compromise between the requirements of empiricists and technological constraints. In this sense, the famous model of circulation's overall control [45] is an exclusive one both by its enormous complexity and by the attempt to test several concepts and hypotheses concerning general human physiology. Studying this model was obligatory work for each modeler thus we should make several notations here.

First of all, the model was not strongly identified. Critics considered this a sufficient reason not to notice or downplay the model's merits. But the main author - Arthur Guyton was a very famous and well- recognized physiologist one to suggest that the quantitative relationships used in the model approximately do correspond to reality. It is worth to underlay that the model incorporated 16 organs and AFS influencing the parameters of CVS and determining both central and regional circulation. Many curves and constants reflecting the current state of each modeled unit were tunable. But authors have had not proposed a reasonable algorithm for automatically tuning: they did it in manual mode, based on their experiences. By the way, authors of another complex model [63], looked like the model of Guyton et al [13] model, were also forced to simulate physiological events in the rest condition or under the gradual exercise by altering constants in manual mode.

To our opinion, the model of Guyton et al [13] had one big aim - to make a theoretic analysis of complex physiological events as it is possible. However, such models also met specific problems concerned technologies of simulations. The model incorporated physiological mechanisms of essentially different dynamics. Some events go on within seconds while others, to show essential alterations, need hours or months. Neither mathematical methods nor computers' power was capable to provide simultaneous modeling. Thus, the authors tried to make partial simulations and then combine their results manually. Despite modern computers exceeding the power of computers of XX Century thousand times, currently, the simultaneously modeling of multi-scale biological transformations is still an actual problem and nowadays.Shim et al [64] have developed a comprehensive cell model that simulates the sequential cellular events from membrane excitation to contraction in the human ventricle. By combining this ventricular cell model with a lumped circulation model, they examined how blood pressure dynamics in the ventricle and aorta are related to the cellular processes. However, to convert cell contraction into ventricular pressure authors have used Laplace's law, introducing a simple geometric model of a ventricle: one shaped like a thin-walled hemisphere. The time courses of the hemodynamic properties, as well as the volume-pressure trajectory of the left ventricle, were well reproduced. The multi-scale cardiovascular model, which covers from cardiac cells to the circulatory system, simulates the typical characteristics of heart mechanics, such as the pressure-volume relationship, stroke volume and the effect of the increased maximum free calcium concentration on cardiovascular hemodynamics. The variation due to different pacing frequencies for myocyte excitation was also investigated to assess the



DOI: https://doi.org/10.30564/jhp.v2i1.1905

effects of heart rate on cardiac cells and the circulatory system.

To model real physiological mechanisms acting on a whole organism-scale, it is necessary to take into account the followings. First of all, the modeler needs a correct conceptual vision of the organs' interaction. Second, he\she should take into account mechanisms providing the self-adaptiveness of the cell. Third, it also is necessary a conceptual vision of mechanisms-enhancers that are activating when the cell-scale mechanisms are not capable of lonely provide cell survival. This triad duly understood should be transferred into a technology in which the cell-scale events are the primary ones while every next-scales' mechanisms represent assistant mechanisms evolutionarily being saved because they enhance the cell capability to provide optimal-like metabolism. Namely, these ideas were proposed and argued in recent publications [31-34,65,66].

Even accepting this recommended ideology, the modeler should not ignore the effects of two other fundamental cell-scale facts. The first one concerns the rate of metabolism. Though its mean value is genetically predetermined in each cell type, the metabolic rate is essentially varying in different phases of the cell cycle. In addition, the duration of every phase is not stable but depends upon material incomes. Therefore, special check-point mechanisms control whether products to be synthesized within the current phase are already present. The second effect mostly expressed in exciting cells, appears because of influences that came from other cells. Namely, due to these stimulating or inhibiting influences, both the integrative physiology and behaviors are provided [34]. However, the integrating influences disrupt the normal course of metabolism, cause internal destruction in the target cell. Therefore, special mechanisms, which can be read in more detail in [31-34], activate mechanisms that serve the synthesis of ATP molecules. As many organs take part in obtaining, transforming, and delivering of source materials for the synthesis of ATP in a cell, they all together are considered functionally to be physiological super-systems (PS). Another PS is providing the chemical purity of the cytoplasm.

To realistically model physiological events in multicellular objects (in a cells' population, in organs built of multiple such populations, as well as in AFS, and PS), the modeler also should take into account dynamic effects caused by internal heterogeneities of cells into the cell population [65].

So, the multi-scale modeling, to be capable of realistically simulating both the normal physiological mechanisms and multiple scenarios leading to the abnormal working of these mechanisms, needs to be based on the recent concept explaining the metabolic and functional integrity of human specialized cells [34].

In the review [67], authors discussing research efforts to the whole-heart modeling. The models seems to be multi-scale are rather describing biophysical than physiological mechanisms. The same is true for the models like [69-71] while models described in [72-74] combine both physiological and biophysical mechanisms. The multi-scale model of the heart, described in [75], illustrates the effects provided by using massively parallel computers. It is undoubtedly that supercomputers opening new opportunities for simulations in physiology.

Clinical models we have not analyzed because they include a lot of nuances specific for each pathology but not principal in the frame of our overview.

It is worth to return to the questions first formulated by Schrödinger in [4] and described above. Three main answers are now known. Tertiary and quaternary macromolecules, which make up most of the biological structures of the cell, are sensitive to thermodynamic and other fluctuations. Therefore, a permanent compensatory synthesis of destroyed molecules takes place in the cell. Fluctuations also affect the throughput of the portal mechanisms of cell membranes, disrupting the resting potential of the cell. Their recovery is possible with the consumption of ATP. In addition to these reasons, in a multicellular organism of animals or humans, the integration of heterogeneous cells during the functioning of organs and systems also contributes to the additional energy expenditure [34]. Thus, it is impossible to correctly model physiological multi-scale processes and phenomena without properly describing the energy dependence of the simulated effects in these models. Since ATP is synthesized from the limited internal resources of carbohydrates and oxygen, in fact, these restrictions significantly modulate both organ and body physiology. So far, only the first attempts have been made to create an integrated model of the physiological megasystem providing the energy balance in each cell of the body [31-34,76]. We believe that this aspect of modeling should be further developed in future models of human physiology and pathophysiology.

## 5. Conclusion

Mathematical modeling now is a legal method of physiological research. Multiple quantitative models describe functions of human organs, anatomical-functional systems (AFS), and functional systems (FS) under norm and certain pathologies. Most models, operating with both measured data and virtual data or heuristics, give the physiologists-empiricists additional information and help them to better understand intimate mechanisms governing


DOI: https://doi.org/10.30564/jhp.v2i1.1905

the living system. As the physiological concepts serve the basis of the models and specialized computer simulators, simulations cannot provide conclusions cardinally differing from the used concept. This is why nether principles or rules organizing and governing the human integrative physiology at rest and/or under behavioral activities have not been clarified yet by simulations. Moreover, even concepts used in most models of organs or AFSs suggest the organ or AFS have its physiological goals and mechanisms for their providing. In fact, at physiological levels of life organization, there is none special goal: it is an illusion born by mechanisms structurally-functionally linked in forms of special negative or positive feed-backs to supply cells by energy and materials in order that makes possible the entire chain to repetitively do biological works. Every mechanism combining reciprocally activating sub-mechanisms damps the extreme oscillations of life variables thus creating the illusion of the local goal. In order to comprehend the principles of such mechanisms at multiple scales, it is necessary to create novel mathematical models (NMM). Cellular self-regulators must be the basis of NMM. As every self-regulator possesses limited power, NMM also must contain models describing conditions for appearing/disappearing of special chemical agents that are penetrating into the blood and activating additional multi-cellular enhancers of intracellular self-regulators. As cells simultaneously fighting for common resources, the entire organism will be modeled as a net of physiological super-systems together supporting cells' optimal physiology. Only such models could simulate both short-term and long-term adaptive responses of the human organism to internal/external destructive challenges.

## References


[1] Cannon WB. Organization for physiological homeostasis. Physiol. Rev., 1929, 9: 399-431.

[2] Wiener N., Rosenblueth A. The mathematical formulation of the problem of conduction of impulses in a network of connected excitable elements, specifically in cardiac muscle. Arch. Inst. Cardiologia de Mexico. 1946, XVI(3-4): 205-265.

[3] Wiener N. Cybernetics: Or Control and Communication in the Animal and the Machine. 1948, Technology Press.

[4] Schrodinger E. What is life? The Physical Aspect of the Living Cell. 1944, Cambridge University Press,

[5] Grodins FS, Gray JS, Schroeder KR, Norins AL, Jones RW. Respiratory responses to CO2 inhalation. A theoretical study of a nonlinear biological regulator. J Appl Physiol. 1954, 7(3): 283-308.

[6] Grodins F. Control Theory and Biological Systems. N. Y.: Columbia. Univ. Press, 1963.

[7] Beneken GW. Investigation on the regulatory system of the blood circulation. In: Circulatory Analog Computers. Amsterdam: North-Holland Publishing company, 1963: 16-28.

[8] Beneken GW. A mathematical approach to cardiovascularfunction. The uncontrolled human system. Drukkerij Elinknjk Utrecht, 1965.

[9] Defares YJ, Osborn JJ, Hara HH. Theoretical synthesis of the cardiovascular system. Acta Physiol, Pharmacol., -Neerl., 1962, 12: 189-265.

[10] De Pater L., van den Berg J.W. An Electrical Analogue of the Entire Human Circulatory System. Medical Electronics and Biological Engineering, 1964, 11: 161-166.

[11] Scher AM, Young AC. Servoanalysis of carotid sinus effects on peripheral resistance. Circ. Res., 1963, 12: 152-162.

[12] Defares Y.J. Examples of cardiovascular models simulation of the cardiovascular system. J. Chron. Dis., 1966, 19: 401-410.

[13] Guyton AC, Coleman TG, Granger HJ. Circulation. Overall Regulation. Annual Review of Physiology, 1972, 34: 13-46.

[14] Sagawa K. Comparative models of overall circulatory mechanics. In: Brown JHV, Dikson JP Advances in Bio-med. Eng. Academ. Press. New York, London, 1973, 3: 1-96.

[15] Sagawa K. Concerning "gain". Am J Physiol., 1978, 235(2): H117.

[16] Guyton AC. Renal servo-control of arterial blood pressure. J. Appl. Physiol, 1967, 22: 139-142.

[17] Guyton AC. Long-term arterial pressure control: an analysis from animal experiments and ... in the pathogenesis of hypertension, N Engl J Med., 1966, 356: 2007.

[18] Osborn JW, Jacob F, Guzman P. A neural set point for the long-term control of arterial pressure: beyond the arterial baroreceptor reflex. Am J Physiol Regul Integr Comp Physiol. 2005, 288(4): R846-855.

[19] Brands MW. Chronic blood pressure control. Compr Physiol. 2012, 2(4): 2481-2494.

[20] Dorrington KL, Pandit JJ. The obligatory role of the kidney in long-term arterial blood pressure control: extending Guyton's model of the circulation. Anaesthesia., 2009, 64(11): 1218-1228.

[21] Nishida Y, Tandai-Hiruma, M, Kemuriyama, T. et al. Long-term blood pressure control: is there a set-point in the brain?. J Physiol Sci. 2012, 62: 147-161. https://doi.org/10.1007/s12576-012-0192-0

[22] BensonJF, SchoemanJP, VenterFJ, Ker JA, ZeilerGE, Bester L, van Niekerk J, Tintinger




DOI: https://doi.org/10.30564/jhp.v2i1.1905

GR. Aortic Arch Baroreceptor Stimulation in an Experimental Goat Model: A Novel Method to Lower Blood Pressure Front. Cardiovasc. Med., 2019. https://doi.org/10.3389/fcvm.2018.00193

[23] Burgoyne S, Georgakopoulos D, Belenkie I, Tyberg JV. Systemic vascular effects of acute electrical baroreflex stimulation. AJP Hear Circ Physiol. 2014, 307:H236-241. DOI: 10.1152/ajpheart.00422.2013

[24] Hori D, Max L, Laflam A, Brown C, Neufeld KJ, Adachi H, Sciortino C, Conte JV, Cameron DE, Hogue CW Jr, Mandal K. Blood Pressure Deviations From Optimal Mean Arterial Pressure During Cardiac Surgery Measured With a Novel Monitor of Cerebral Blood Flow and Risk for Perioperative Delirium: A Pilot Study. J Cardiothorac Vasc Anesth. 2016, 30(3): 606-612. DOI: 10.1053/j.jvca.2016.01.012

[25] Höcht C. Blood Pressure Variability: Prognostic Value and Therapeutic Implications. International Scholarly Research Notices, Notices, 2013. https://doi.org/10.5402/2013/398485

[26] Raven PB, Chapleau MW. Blood pressure regulation: overview and future research directions. Eur J Appl Physiol. 2014, 114(3): 579-586. DOI: 10.1007/s00421-014-2823-z

[27] Nishi EE, Bergamaschi CT, Campos RR. The crosstalk between the kidney and the central nervous system: the role of renal nerves in blood pressure regulation. Exp Physiol. 2015, 100(5): 479-484. DOI: 10.1113/expphysiol.2014.079889

[28] Averina VA, Othmer HG, Fink GD, Osborn JW. A new conceptual paradigm for the haemodynamics of salt-sensitive hypertension: a mathematical modelling approach. J Physiol. 2012, 590(23): 5975-5992.

[29] Bolívar JJ. Essential Hypertension: An Approach to Its Etiology and Neurogenic Pathophysiology. Int J Hypertens. 2013. DOI: 10.1155/2013/547809

[30] Ivy JR, Bailey MA. Pressure natriuresis and the renal control of arterial blood pressure. J Physiol. 2014, 15, 592(18): 3955-67. DOI: 10.1113/jphysiol.2014.271676

[31] Grygoryan R.D. Comprehension of individual adaptation mechanisms: endogenous tuning of constants determining optimal physiological states. Slovak international scientific journal, 2019, 32: 67-72.

[32] Grygoryan R.D., Sagach V.F. The concept of physiological supersystems: New stage of integrative physiology. Intern. J. of Physiol. and Pathophysiology, 2018, 9(2): 169-180.

[33] Grygoryan RD. Endogenous variators of individual health corridor. Znanstvena misel journal, 2019, 35: 31-37.

[34] Grygoryan RD. The Optimal Coexistence of Cells: How Could Human Cells Create The Integrative Physiology. Journal of Human Physiology.2019, 1 (01):8-28. DOI: 10.30564/jhp.v1i1.1386

[35] Kokalari I. Review on lumped parameter method for modeling the blood flow in systemic arteries. Journal of Biomedical Science and Engineering 2013, 06(01): 92-99. DOI: 10.4236/jbise.2013.61012

[36] Shimizu S. Une D, KawadaT, HayamaY, KamiyaA, Shishido T, Sugimachi T. Lumped parameter model for hemodynamic simulation of congenital heart diseases The Journal of Physiological Sciences, 2018, 68: 103-111. https://doi.org/10.1007/s12576-017-0585-1.

[37] Corsini C, Baker C, Kung E, Schievano S, Arbia G, Baretta A, Biglino G, Migliavacca F, Dubini G, Pennati G, Marsden A, Vignon-Clementel I, Taylor A, Hsia TY, Dorfman A. Modeling of Congenital Hearts Alliance (MOCHA) Investigators: An integrated approach to patient-specifc predictive modeling for single ventricle heart palliation. Comput Methods Biomech Biomed Engin., 2014, 17: 1572-1589.

[38] Dampney RAL. Resetting of the Baroreflex Control of Sympathetic Vasomotor Activity during Natural Behaviors: Description and Conceptual Model of Central Mechanisms. Front Neurosci., 2017, 11: 461. Published 2017. DOI: 10.3389/fnins.2017.00461

[39] Wallin BG, Charkoudian N. Sympathetic neural control of integrated cardiovascular function: insights from measurement of human sympathetic nerve activity. Muscle Nerve. 2007, 36(5): 595-614.

[40] Pope SR, Ellwein L, Zapata EC, Novak V., Kelley CT, Olufsen MS. Estimation and identification of parameters in a lumped cerebrovascular model, Mathematical Biosciences and Engineering 2009, 6(1): 93-115.

[41] Rideout V. Mathematical and computer modeling of physiological systems, Prentice Hall, Englewood Cliffs, NJ, 1991.

[42] Sunagawa K, Sagawa K. Models of ventricular contraction based on time-varying elastance, Critical Revies in Biomedical Engineering, 1982, 7(3): 193-228.

[43]Timischl S. A global model of the cardiovascular and respiratory system, Ph.D. thesis, University of Graz, Institute for Mathematics and Scientific Computing, 1998.

[44] Grygoryan RD, Degoda AG, Dzhurinsky EA, Khar-

 DOI: https://doi.org/10.30564/jhp.v2i1.1905

sun DS. A simulator of a pulsatile heart. Problrms in programming, 2017, 4: 98-108.
DOI: https://doi.org/10.15407/pp2017.04.098
[45] Cowley A. W., Jr. Long-term control of arterial blood pressure. Physiol Rev., 1992, 72: 231-300.
[46] The IUPS Physiome Project. http://www.iups.org/physiome-project/
[47] Hunter PJ, Viceconti M. The VPH-Physiome Project: Standards and Tools for Multiscale Modeling in Clinical Applications. 2009 IEEE Reviews in Biomedical Engineering, 2: 40-53. DOI: 10.1109/RBME.2009.2036204
[48] Bassingthwaighte JB and Chinn TM. Re-examining Michaelis-Menten enzyme kinetics for xanthine oxidase. Adv Physiol Educ, 2013, 37: 37-48.
[49] Bassingthwaighte JB, Butterworth E, Jardine B, Raymond G. Compartmental modeling in the analysis of biological systems. In: Computational Toxicology (Methods in Molecular Biology Series), edited by Brad Reisfeld and Arthur N Mayeno. New York NY: Springer Science+Business Media LLC, 2012, 929: 391-438.
[50] Bassingthwaighte JB, Beard DA, Carlson BE, Dash RK, Vinnakota K. Modeling to link regional myocardial work, metabolism and blood flows. Ann Biomed Eng. 2012, 40(11): 2379-98. DOI: 10.1007/s10439-012-0613-5
[51] Niederer FM, Cherry EM, Fenton FH, Koivumäki JT, Seemann G, Thul R, Zhang H, Sachse FB, Beard D, et al. Prog Biophys Mol Biol., 2011, 104(1-3): 2-21.
[52] Maas AH, Rozendaal YJ, van Pul C, Hilbers PA, Cottaar WJ, Haak HR, van Riel NA. A physiology-based model describing heterogeneity in glucose metabolism: the core of the Eindhoven Diabetes Education Simulator (E-DES). J Diabetes Sci Technol., 2015, 9(2): 282-292.
[53] Shiang KD, Kandeel F. A computational model of the human glucose-insulin regulatory system. J Biomed Res., 2010, 24(5): 347-364. DOI:10.1016/S1674-8301(10)60048-6
[54] De Gaetano A, Gaz C, Panunzi S. Consistency of compact and extended models of glucose-insulin homeostasis: The role of variable pancreatic reserve. PLoS ONE, 2019. https://doi.org/10.1371/journal.pone.0211331
[55] González AA, Voos H, Darouach M. Glucose-Insulin System Based on Minimal Model: A Realistic Approach. 2015 17th UKSim-AMSS International Conference on Modelling and Simulation (UKSim), Cambridge, 2015, 55-60. DOI: 10.1109/UKSim.2015.65
[56] Grygoryan R.D. High sustained G-tolerance model development.STCU#P-078 EOARD# 01-8001 Agreement: Final Report, 2002.
[57] Grygoryan RD, Hargens AR. A virtual multicellular organism with homeostatic and adaptive properties. In: Adaptation Biology and Medicine: Health Potentials. Ed. L. Lukyanova, N.Takeda, P.K. Singal. New Delhi: Narosa Publishing House, 2008, 5: 261-282.
[58] White RJ, Leonard JI, Rummel JA, Leach CS. A systems approach to the physiology of weightlessness. Journal of medical systems, 1982, 6(4): 343-358.
[59] Hensley DW1, Mark AE, Abella JR, Netscher GM, Wissler EH, Diller KR. 50 years of computer simulation of the human thermoregulatory system. J Biomech Eng., 2013, 135(2): 021006. DOI: 10.1115/1.4023383
[60] Alzeer AH, Wissler EH. Theoretical analysis of evaporative cooling of classic heat stroke patients. Int J Biometeorol. 2018, 62(9): 1567-1574. DOI: 10.1007/s00484-018-1551-1
[61] Wagar LE, DiFazio RM. Davis MM. Advanced model systems and tools for basic and translational human immunology. Genome Med. 2018, 10(73). https://doi.org/10.1186/s13073-018-0584-8.
[62] Kerepesi C, Bakács T, Szabado, T. MiStImm: an agent-based simulation tool to study the self-nonself discrimination of the adaptive immune response. Theor Biol Med Model, 2019, 16(9). https://doi.org/10.1186/s12976-019-0105-5
[63] Amosov NM, Paleys BL, Agapov BT, Yermakova II, Lyabakh KG, Solovev AA. Theoretical Investigations of Human Physiology. Kiev, Naukova Duma, 1977.
[64] Shim EB, Leem CH, Abe Y, Noma A. A new multiscale simulation model of the circulation: from cells to system. Philos Trans A Math Phys Eng Sci. 2006, 364(1843): 1483-1500. https://doi.org/10.1098/rsta.2006.1782
[65] Grygoryan RD. The Energy basis of reversible adaptation. N.Y.: Nova Science, 2012: 254. ISBN: 978-1-62081-093-4
[66] Grygoryan RD. The optimal circulation: cells contribution to arterial pressure. N.Y.: Nova Science, 2017: 287. ISBN: 978-1-53612-295-4
[67] Trayanova NA, Winslow R. Whole-Heart Modeling. Applications to Cardiac Electrophysiology and Electromechanics. Circulation Research. 2011, 108: 113-128. https://doi.org/10.1161/CIRCRESAHA.110.223610
[68] Noble D, Rudy Y. Models of cardiac ventricular action potentials: iterative interaction between experiment and simulation. Philos Trans R Soc Lond A. 2001, 359: 1127-1142.



DOI: https://doi.org/10.30564/jhp.v2i1.1905

[69] Rice JJ, Wang F, Bers DM, de Tombe PP. Approximate model of cooperative activation and crossbridge cycling in cardiac muscle using ordinary differential equations. Biophys J., 2008, 95: 2368-2390.

[70] Niederer SA, Hunter PJ, Smith NP. A quantitative analysis of cardiac myocyte relaxation: a simulation study. Biophys J. 2006, 90: 1697-1722.

[71] Trayanova NA, Winslow R. Whole-Heart Modeling: Applications to Cardiac Electrophysiology and Electromechanics. Circulation Research. 2011, 108: 113-128.
https://doi.org/10.1161/CIRCRESAHA.110.223610

[72] Kerckhoffs RC, Neal ML, Gu Q, Bassingthwaighte JB, Omens JH, McCulloch AD. Coupling of a 3D finite element model of cardiac ventricular mechanics to lumped systems models of the systemic and pulmonic circulation. Ann Biomed Eng., 2007, 35: 1-18.
DOI: 10.1007/s10439-006-9212-7

[73] Walpole J, Papin JA, Peirce SM. Multiscale computational models of complex biological systems. Annu Rev Biomed Eng. 2013, 15: 137-154.
DOI: 10.1146/annurev-bioeng-071811-150104

[74] Jie X, Gurev V, Trayanova N. Mechanisms of mechanically induced spontaneous arrhythmias in acute regional ischemia. Circ Res. 2010, 106: 185-192.
https://doi.org/10.1161/CIRCRESAHA.109.210864

[75] A. Hosoi, T. Washio, J. Okada, Y. Kadooka, K. Nakajima, T. Hisada. A Multi-Scale Heart Simulation on Massively Parallel Computers. SC'10: Proceedings of the 2010 ACM/IEEE International Conference for High Performance Computing, Networking, Storage and Analysis, New Orleans, LA, 2010, 1-11.
DOI: 10.1109/SC.2010.5

[76] Grygoryan R.D., Aksenova T.V., Degoda A.G. A simulator of mechanisms providing energy balance in human cells. Cybernetics and Computing Technologies. 2017, 2: 67-76.
DOI: https://doi.org/10.15407/kvt188.02.065

 DOI: https://doi.org/10.30564/jhp.v2i1.1905

# Author Guidelines

This document provides some guidelines to authors for submission in order to work towards a seamless submission process. While complete adherence to the following guidelines is not enforced, authors should note that following through with the guidelines will be helpful in expediting the copyediting and proofreading processes, and allow for improved readability during the review process.

## Ⅰ. Format

- Program: Microsoft Word (preferred)
- Font: Times New Roman
- Size: 12
- Style: Normal
- Paragraph: Justified
- Required Documents

## Ⅱ. Cover Letter

All articles should include a cover letter as a separate document.

The cover letter should include:

- Names and affiliation of author(s)

The corresponding author should be identified.

Eg. Department, University, Province/City/State, Postal Code, Country

- A brief description of the novelty and importance of the findings detailed in the paper

Declaration

v Conflict of Interest

Examples of conflicts of interest include (but are not limited to):

- Research grants
- Honoria
- Employment or consultation
- Project sponsors
- Author’s position on advisory boards or board of directors/management relationships
- Multiple affiliation
- Other financial relationships/support
- Informed Consent

This section confirms that written consent was obtained from all participants prior to the study.

- Ethical Approval

Eg. The paper received the ethical approval of XXX Ethics Committee.

- Trial Registration

Eg. Name of Trial Registry: Trial Registration Number

- Contributorship

The role(s) that each author undertook should be reflected in this section. This section affirms that each credited author has had a significant contribution to the article.

1. Main Manuscript
2. Reference List
3. Supplementary Data/Information

Supplementary figures, small tables, text etc.

As supplementary data/information is not copyedited/proofread, kindly ensure that the section is free from errors, and is presented clearly.

## Ⅲ. Abstract

A general introduction to the research topic of the paper should be provided, along with a brief summary of its main results and implications. Kindly ensure the abstract is self-contained and remains readable to a wider audience. The abstract should also be kept to a maximum of 200 words.

Authors should also include 5-8 keywords after the abstract, separated by a semi-colon, avoiding the words already used in the title of the article.

Abstract and keywords should be reflected as font size 14.

## Ⅳ. Title

The title should not exceed 50 words. Authors are encouraged to keep their titles succinct and relevant.

Titles should be reflected as font size 26, and in bold type.

## Ⅳ. Section Headings

Section headings, sub-headings, and sub-subheadings should be differentiated by font size.

Section Headings: Font size 22, bold type

Sub-Headings: Font size 16, bold type

Sub-Subheadings: Font size 14, bold type

Main Manuscript Outline

## Ⅴ. Introduction

The introduction should highlight the significance of the research conducted, in particular, in relation to current state of research in the field. A clear research objective should be conveyed within a single sentence.

## Ⅵ. Methodology/Methods

In this section, the methods used to obtain the results in the paper should be clearly elucidated. This allows readers to be able to replicate the study in the future. Authors should ensure that any references made to other research or experiments should be clearly cited.

## Ⅶ. Results

In this section, the results of experiments conducted should be detailed. The results should not be discussed at length in

this section. Alternatively, Results and Discussion can also be combined to a single section.

## Ⅷ. Discussion

In this section, the results of the experiments conducted can be discussed in detail. Authors should discuss the direct and indirect implications of their findings, and also discuss if the results obtain reflect the current state of research in the field. Applications for the research should be discussed in this section. Suggestions for future research can also be discussed in this section.

## Ⅸ. Conclusion

This section offers closure for the paper. An effective conclusion will need to sum up the principal findings of the papers, and its implications for further research.

## Ⅹ. References

References should be included as a separate page from the main manuscript. For parts of the manuscript that have referenced a particular source, a superscript (ie. [x]) should be included next to the referenced text.

[x] refers to the allocated number of the source under the Reference List (eg. [1], [2], [3])

In the References section, the corresponding source should be referenced as:

[x] Author(s). Article Title [Publication Type]. Journal Name, Vol. No., Issue No.: Page numbers. (DOI number)

## Ⅺ. Glossary of Publication Type

J = Journal/Magazine

M = Monograph/Book

C = (Article) Collection

D = Dissertation/Thesis

P = Patent

S = Standards

N = Newspapers

R = Reports

Kindly note that the order of appearance of the referenced source should follow its order of appearance in the main manuscript.

Graphs, Figures, Tables, and Equations

Graphs, figures and tables should be labelled closely below it and aligned to the center. Each data presentation type should be labelled as Graph, Figure, or Table, and its sequence should be in running order, separate from each other.

Equations should be aligned to the left, and numbered with in running order with its number in parenthesis (aligned right).

## Ⅻ. Others

Conflicts of interest, acknowledgements, and publication ethics should also be declared in the final version of the manuscript. Instructions have been provided as its counterpart under Cover Letter.

## About the Publisher

Bilingual Publishing Co. (BPC) is an international publisher of online, open access and scholarly peer-reviewed journals covering a wide range of academic disciplines including science, technology, medicine, engineering, education and social science. Reflecting the latest research from a broad sweep of subjects, our content is accessible world-wide——both in print and online.

BPC aims to provide an analytics as well as platform for information exchange and discussion that help organizations and professionals in advancing society for the betterment of mankind. BPC hopes to be indexed by well-known databases in order to expand its reach to the science community, and eventually grow to be a reputable publisher recognized by scholars and researchers around the world.

BPC adopts the Open Journal Systems, see on ojs.bilpublishing.com

# Database Inclusion

Asia & Pacific Science Citation Index

Creative Commons

China National Knowledge Infrastructure

Google Scholar

Crossref

MyScienceWork

BILINGUAL
PUBLISHING CO.
Pioneer of Global Academics Since 1984

Tel:+65 65881289
E-mail:contact@bilpublishing.com
Website:www.bilpublishing.com