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# What is in common between preeclampsia, HPS70 and medieval headwear? Part I. Serum HPS70 in preeclampsia: systematic review and meta-analysis

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***The objective:*** **to investigate the relationship between HSP70 concentrations in maternal serum and preeclampsia and assess the prospects of using HSP70 as a preeclampsia predictor.**
***Materials and methods*****. The original publications, which study HSP70 in maternal serum of preeclamptic women, were searched and analyzed. Papers were identified with Scopus, PubMed Central, Virtual Health Library databases, published before January 2023, the keywords were «HSP70», «preeclampsia», «heat shock protein 70», «pregnant». Statistical analysis was performed via software EZR 1.55.**
***Results*****. 16 case-control studies were included, making a total of 751 pregnant women with preeclampsia and 719 healthy pregnant women. The analysis found the statistically significant difference between HSP70 concentrations in maternal serum of preeclamptic and healthy pregnant patients. Cochrane Q-test showed high heterogeneity among studies ($p<0.01$), the value of the $I^2$ statistic was 97%.**
**Dividing the studies into groups made it possible to reduce or remove heterogeneity completely. This high level of heterogeneity for publications together, but low within most groups, suggests that there are certain factors that significantly influence some studies.**
***Conclusions*****. The conducted systematic review and meta-analysis confidently indicate an increased average serum concentration of HSP70 in pregnant women with preeclampsia compared to healthy pregnant women at the corresponding gestational age.**
**No statistically significant relationship was found between increased HSP70 concentration in preeclampsia and pregnant women's age, gestational age, systolic and diastolic blood pressure. Quantitative assessment of HSP70 levels is complicated by the lack of a single standard for laboratory diagnostics. The case-control design of the presented studies limits their significance.**
**The use of HSP70 as a predictor of preeclampsia is promising, but requires further study and prospective cohort studies.**
***Keywords:*** *HSP70, heat shock protein 70, preeclampsia, chronic hypertension, superimposed preeclampsia, pregnancy, systematic review, meta-analysis.*

## Що спільного між прееклампсією, HSP70 та середньовічним капелюхом? Частина І. Сироватковий HSP70 при прееклампсії: систематичний огляд та мета-аналіз

***О. К. Попель, Д. О. Говсєєв***

***Мета дослідження:*** встановлення зв'язку між концентраціями HSP70 в сироватці крові вагітної та прееклампсією, оцінювання перспективи використання HSP70 у якості предиктора прееклампсії.
***Матеріали та методи.*** Проведені пошук та аналіз оригінальних досліджень, які присвячені вивченню HSP70 у сироватці крові у жінок із прееклампсією. Роботи ідентифікували за допомогою баз Scopus, PubMed Central, Virtual Health Library, пошук включав публікації до січня 2023 року, застосовували ключові слова «HSP70», «preeclampsia», «heat shock protein 70», «pregnant».
Статистичний аналіз проводили за допомогою програмного забезпечення EZR 1.55.
***Результати.*** Було відібрано 16 досліджень типу «випадок-контроль», які сумарно включали 751 вагітну із прееклампсією та 719 здорових вагітних. Проведений аналіз знайшов статистично значущу різницю між сироватковими концентраціями HSP70 у здорових вагітних та вагітних з прееклампсією. Q-тест Кохрена продемонстрував високу гетерогенність серед досліджень ($p<0,01$), значення статистики $I^2$ дорівнювало 97%.
Поділ досліджень на групи дозволив зменшити або виключити гетерогенність. Такий високий рівень гетерогенності для публікацій разом, але невисокий всередині більшості груп, свідчить про те, що існують певні фактори, які істотно впливають на деякі дослідження.
***Висновки.*** Проведені систематичний огляд та мета-аналіз впевнено свідчать про підвищену у середньому сироваткову концентрацію HSP70 у вагітних із прееклампсією порівняно зі здоровими вагітними у відповідний гестаційний термін. Не було виявлено статистично значущої залежності між підвищенням концентрації HSP70 при прееклампсії та віком вагітних, гестаційним терміном, систолічним та діастолічним тиском. Кількісне оцінювання рівнів HSP70 ускладнено





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відсутністю єдиного стандарту лабораторної діагностики. Планування представлених досліджень у вигляді «випадок-контроль» обмежує їхнє значення.
Використання HSP70 у якості предиктора прееклампсії перспективне, але потребує подальшого вивчення та проведення проспективних когортних досліджень.
***Ключові слова:*** *HSP70, білок теплового шоку 70, прееклампсія, хронічна гіпертензія, накладена прееклампсія, вагітність, систематичний огляд, мета-аналіз.*

Preeclampsia remains one of the leading causes of maternal and perinatal morbidity and mortality worldwide [1]. The pathogenesis and predictors of preeclampsia are still under investigation. Preeclampsia even earned epithet the «disease of theories» [2]. One of the areas of preeclampsia research is the study of heat shock proteins (HSP) – proteins whose expression increases under stress factors influence [3–5].

It is believed that the initiating factor of preeclampsia is cytotrophoblast invasion violation and spiral artery remodeling [2, 6, 7]. Placental ischemia development leads to an imbalance between anti-angiogenic and angiogenic factors, generalized endothelial dysfunction, excessive inflammatory response and oxidant stress [6–11]. These conditions, as well as hemodynamic stress, are capable of inducing HSP70 expression [4, 5].

HSP70 is the traditional protein's term, named by mass in daltons. Since 2008, according to Human Genome Organization (HUGO) Gene Nomenclature Committee recommendations, the HSP70 protein family has been named HSPA [12]. It is currently known that the human genome encodes about 15 proteins of the HSPA family (HSP70) [13]. Coding by different genes enables the rapid synthesis of a sufficient number of HSPs in response to many factors and reflects evolutionary diversity [13].

HSP proteins are universal chaperones (from French chaperon – «to accompany», «companion»), that is, proteins that bind to other proteins and perform a number of functions. The term «chaperone» became generally accepted in 1987 after R. John Ellis put forward the hypothesis of universal proteins that are responsible for peptides shape [14]. This name, due to its vividness and imagery, gained widespread popularity.

In the Middle Ages in France, a chaperone was a headwear. The famous French folk tale «Little Red Riding Hood» is called «Le Petit Chaperon rouge». Today we know that all eukaryotic cells have «protective hats» – chaperones.

HSP70 (HSPA1A) is located in the nucleus and cytoplasm of the cell in a complex with the HSP gene transcription factor called heat shock factor (HSF) [15, 16]. When stress factors appear, HSF separates from HSP70, accumulates in the nucleus and activates the production of new HSP70 [15, 16]. HSP70 is an adenosine triphosphate-dependent (ATP-dependent) protein that requires co-factors for activation, one of which is HSP40 [15, 16]. The attachment of HSP40 to HSP70 initiates the hydrolysis of adenosine triphosphate (ATP) to adenosine diphosphate (ADP), which increases the HSP70 affinity with peptides [15, 16]. HSP70 genes expression occur quickly after stress factors occurrence [15, 16]. After the end of the stress factor influence, HSF again joins free HSP70 [15, 16].

HSP70 is involved in many processes of protein's life: folding (spatial folding of the protein molecule), refolding (correction of the incorrect conformation of proteins), translocation through membranes, aggregation or disaggregation, proteolytic degradation of proteins that couldn't be corrected, and is also a component of the apoptosis regulatory mechanism at all its stages [13, 15–19].

HSP70 has also been identified on cell's surface, in the intercellular space, and in blood serum [13, 20]. In the case of extracellular location, HSP70 has additional functions [21]. HSP70 participates in cell cycle, inflammatory and immune reactions [22–24]. HSP70 stimulates the immune response, possibly due to the cross-homology between human and infectious agent's HSP70 [25].

Extracellular HSP70 can act as cytoprotector: for example, under hemodynamic stress influence, it binds to vascular smooth muscle cells surface protecting cells from apoptosis [26]. HSP70 takes part in inflammatory and immune processes: HSP70 binds to CD14, CD91, Toll receptors on antigen-presenting cells and stimulates cytokines production (tumor necrosis factor, interleukin) [27]. Autoimmune reactivity associated with HSP70 can stem from response to peptides, which are produced during cell damage [28, 29].

HSP70 was detected in blood serum of non-pregnant and pregnant women alike [30–32]. Is there an association between serum HSP70 and preeclampsia? Are there any additional factors during pregnancy that may affect serum HSP70 levels? Does HSP70 concentration in pregnant women with superimposed preeclampsia has any peculiarities?

There is a systematic review and meta-analysis Saghafi N. et al. (2018), which studied HSP70 in preeclampsia and included 7 publications from 2002 to 2011 years [4]. However, the question of HSP70 and other HSPs contribution to preeclampsia development has not been settled yet, searches continue, new original studies appear to this day. Publications report HSP concentrations not only in blood serum but also in the placenta, umbilical cord blood, uterine tissues, they may also report the level of gene expression [3, 33, 34]. There are articles whose results suggest a relationship between HSP and hypertension development [18]. Curiosity about HSPs and their role in various processes is only growing. For instance, in response to the query «HSP» the PubMed database gives 107 results among meta-analyses and systematic reviews and more than 17 000 publications of all types. Considering papers published only last 5 years these numbers are 65 and about 4 000, respectively.

The first part of this study focuses on HSP70 research in preeclampsia. For the meta-analysis, it was decided not to be limited by studies of recent years, but to pick relevant articles published anytime to get the most complete picture.

## MATERIALS AND METHODS

### Identification of studies for meta-analysis

A search and analysis of original publications, related to the study of HSP70 level in blood serum in women with

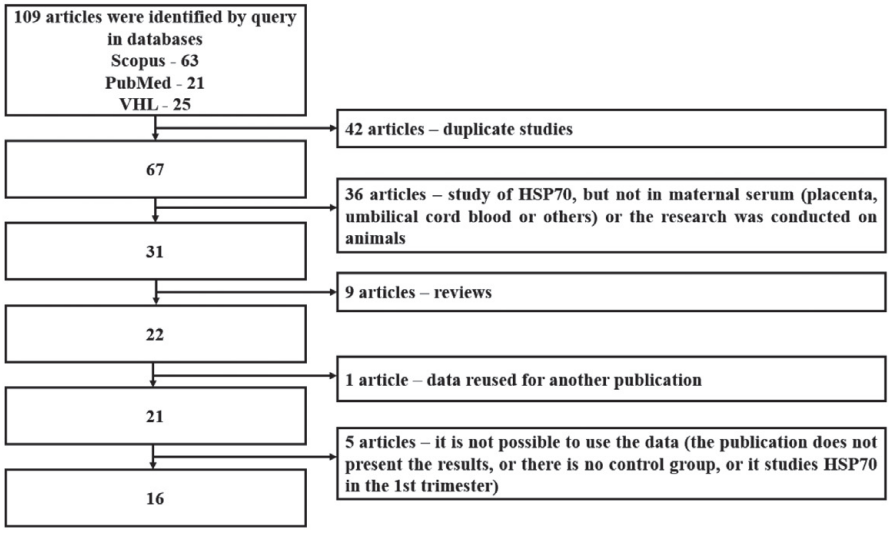


**Figure 1. Flowchart which reflects the selection process**

preeclampsia, was carried out. The meta-analysis was conducted according to the recommendations of PRISMA (2020) [35].

Publications were identified by two researchers independently via Scopus, PubMed Central, Virtual Health Library (VHL) databases, the search included publications up to January 2023. Scopus was searched among «Title, Abstract, Keywords», the query consisted of «HSP70», «heat shock protein 70», «preeclampsia» connected by the AND operator. The query for PubMed and VHL was «HSP70 AND heat AND shock AND protein 70 AND preeclampsia AND pregnant» (for VHL, the search settings was «Title, Abstract, Subject»). The search had no language restrictions.

For every identified by search article the full texts were obtained, reviewed and it was decided whether to include them in the analysis or not. For the meta-analysis, original studies which reported the level of HSP70 in the blood serum of pregnant women and with a cohort or «case-control» design were selected. Exclusion criteria were duplicate articles, reviews, case reports. Studies were included in the analysis if there was reported: sample sizes, serum HSP70 concentrations after 20 weeks in the format of mean with standard deviation (SD) or median with interquartile range (Q1-Q3 – IQR), or median with minimum and maximum values (min-max). The process of selecting publications for meta-analysis is shown in Figure 1.

For meta-analysis were used the results of studies of pregnant women with preeclampsia and control groups of healthy pregnant women. Several researches also looked at pregnant women with gestational hypertension (2 studies – 59 participants), superimposed preeclampsia (1 study – 20 participants), HELLP syndrome (1 study – 10 participants), preeclampsia and fetal growth restriction (FGR) (1 study – 25 participants), active labor at the time of obtaining blood samples (1 study – 50 participants); these subgroups were not included in the meta-analysis to remove additional factors, which may influence result, and create a homogeneous sample.

***Statistical methods***

To apply statistical methods of meta-analysis, the research results were converted into a single format: m±SD, where m – mean, SD – standard deviation. For studies where median and interquartile range or median with minimum and maximum values were reported, the mean and standard deviation were estimated by the Box-Cox transformation method [36], using «bc.mean.sd» function of «estmeansd» library of the R programming language [37].

Standardized mean difference (SMD) was chosen as effect size metric – the difference between the averages of preeclampsia group and control group divided by pooled standard deviation. SMD does not change if studies are conducted in different measurement systems. If the differences in mean values make up the same proportion of the standard deviation, the SMD indicator will be similar [38]. Further analysis was performed in EZR 1.55 software.

A forest plot was built to assess data heterogeneity and clarify the SMD indicator. Publications were divided into 5 groups based on SMD size. Forest plot shows the interval estimates by both common-effects model and random-effects model for every group.

Confidence intervals were calculated using Hedges' G method, weights in forest plot were calculated via inverse variance method. The presence of heterogeneity was checked by the Cochrane Q test, where the restricted maximum-likelihood estimator was used to evaluate $\tau^2$. The $I^2$ statistics was calculated on the basis of Cochrane Q test.

To find the main sources of heterogeneity in the data on HSP70 concentration, a meta-regression model was built (dependency of SMD on other parameters). It is generally recommended to build a meta-regression model only if more than ten studies with the specified parameter [39]. Such criteria were met by maternal age (14 studies), systolic and diastolic pressure (13 studies), gestational age (14 studies). Each parameter was taken in turn as an independent variable in a one-parameter linear regression


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model. However, data on the parameters were reported by the researchers separately for the control and preeclampsia groups. Hence, for meta-regression, the average of the whole sample $\tilde{m}$ was used, which included both control and preeclampsia groups, according to the formula:

$$\tilde{m} = \frac{n_1 \cdot m_1 + n_2 \cdot m_2}{n_1 + n_2}$$

where $n_1$, $n_2$ – participants number in control and preeclampsia groups, respectively;

$m_1$, $m_2$ – average HSP70 number in control and preeclampsia groups, respectively.

The conclusion about the dependence of SMD on parameters (maternal age, systolic and diastolic pressure, gestational age) was drawn based on the statistical significance of linear regression coefficients.

The presence of publication bias was checked via funnel plot, Egger's statistical test was used, with standard deviation as a predictor, weights were calculated using inverse variance. Additionally, the presence of publication bias was checked by DOI plot and calculating the Lewis-Furuya-Kanamori asymmetry index (LFK index) [40]. The DOI graph and LFK index were calculated using «lfkindex» function of «metasens» library of the R programming language.

Sensitivity analysis also was conducted via leave-one-out method. The function «metainf» of «meta» library of the R programming language was used for this purpose.

## RESULTS

As a result of a database search, 109 publications were found. After excluding articles that were duplicated or did not meet the inclusion criteria, 16 original studies were selected for analysis (Figure 1), making a total of 751 women with preeclampsia and 719 healthy pregnant women.

Data on HSP70 concentrations in preeclampsia and control groups were extracted from 16 studies. If required values were presented only on plot, it was extracted via online service WebPlotDigitalizer. Numerical data of 8 studies are translated into the format m±SD, where $m$ – average, SD – standard deviation. Table 1 shows selected studies, reported HSP70 concentrations (if necessary, converted to mean and standard deviation format), number of participants.

It is noticeable from Table 1, that the average level of HSP70 varied significantly among studies, sometimes by one order of magnitude or even two. Therefore, for effect size measurement in the meta-analysis, the standardized mean difference (SMD) was chosen. A forest plot was built and included studies were divided into 5 groups depending on the size of the SMD (Figure 2). It is necessary to note that Zhu J. et al. 2014 [34] contained data on HSP70 concentration separately for preeclampsia and severe preeclampsia groups. Therefore, it was included in the meta-analysis as 2 separate studies: one with calculated SMD for preeclampsia versus controls, the other for severe preeclampsia versus controls. Thus, the control group for Zhu J. et al. 2014 [34] was included in the meta-analysis twice. That's why forest plot on Figure 2 shows a total of 749 healthy pregnant women.

When building forest plot the following groups were identified:

- Group A – articles, where SMD is in 0.81 – 1.39 interval;
- Group B – articles, where SMD is in -0.06 – 0.30 interval;
- Group C – articles, where SMD is in 12.10 – 13.28 interval;
- Group D – articles, where SMD is in 2.50 – 6.36 interval;
- Group E – articles, where SMD was -2.57.

For A, B, C, D groups separate forest plots were built (respectively, fig 2a, 2b, 2c, 2d) for more convenient data visualization.

*Table 1*

**Studies, included in meta-analysis, reported HSP70 concentrations and converted to mean and standard deviation format, number of participants**

| Publication | Control group | | | Preeclampsia group | | |
|---|---|---|---|---|---|---|
| | HSP70, ng/ml, reported in article | HSP70, ng/ml, converted | Sample size | HSP70, ng/ml, reported in article | HSP70, ng/ml, converted | Sample size |
| Jirecek S. et al. 2002 [41] | 1.010 ± 1.380 | – | 55 | 2.820 ± 8.330 | – | 55 |
| Livingston J. et al. 2002 [42] | 30.100 ± 11.500 | – | 51 | 35.400 ± 96.700 | – | 47 |
| Fukushima A. et al. 2005 [43] | 6.100 ± 0.600 | – | 46 | 24.400 ± 3.600 | – | 7 |
| Molvarec A. et al. 2006 [5] | 0.310 (0.270 – 0.390)* | 0.332 ± 0.094 | 127 | 0.550 (0.420 – 0.800)* | 0.646 ± 0.328 | 93 |
| Molvarec A. et al. 2007 [44] | 0.3 (0.270 – 0.330)* | 0.262 ± 0.057 | 20 | 0.540 (0.470 – 0.790)* | 0.625 ± 0.334 | 20 |
| Molvarec A. et al. 2009 [45] | 0.280 (0.030 – 0.590)** | 0.283 ± 0.115 | 70 | 0.580 (0.150 – 3.470)** | 0.726 ± 0.525 | 67 |
| Molvarec A. et al. 2011 [46] | 0,280 (0.210 – 0.310)* | 0.299 ± 0.047 | 60 | 0,580 (0.390 – 0.810)* | 0.639 ± 0.276 | 60 |
| Saghafi N. et al. 2013 [47] | 0 (0 – 4.000)* | 2.716 ± 0.693 | 39 | 9.000 (0 – 23.500)* | 9.424 ± 6.843 | 41 |
| Zhu J. et al. 2014 [34] | 1.880 ± 0.790 | – | 30 | 2.610 ± 0.980 | – | 30 |
| | | | | 3.100 ± 1.180 | – | 30 |
| Akbarzadeh-Jahromi M. et al. 2015 [48] | 0.763 ± 0.091 | – | 31 | 0.504 ± 0.107 | – | 31 |
| Romao-Veiga M. et al. 2018 [49] | 0.680 (0.008 – 1,090)** | 0.668 ± 0.216 | 20 | 0.907 (0.405 – 1.273)** | 0.885 ± 0.221 | 20 |
| Álvarez-Cabrera M. 2018 [50] | 1.600 ± 0.120 | – | 28 | 2.338 ± 0.113 | – | 62 |
| Zhou X. et al. 2019 [51] | 36.000 ± 0.070 | – | 40 | 3.920 ± 0.350 | – | 86 |
| Hua Lai et al. 2020 [52] | 0.480 ± 0.220 | – | 50 | 3.230 ± 1.760 | – | 30 |
| Romao-Veiga M. et al. 2020 [53] <34 week | 0.800 (0.500 – 0.900)** | 0.774 ± 0.102 | 16 | 4.500 (0.800 – 6.000)** | 4.360 ± 0.930 | 26 |
| Romao-Veiga M. et al. 2020 [53] >34 week | 0.700 (0.600 – 0.900)** | 0.717 ± 0.082 | 16 | 0.600 (0.090 – 1.900)** | 0.693 ± 0.471 | 26 |
| Romao-Veiga M. et al. 2022 [54] | 0.674 (0,008 – 1,082)** | 0.664 ± 0.227 | 20 | 0.901 (0.402 – 1.264)** | 0.889 ± 0.212 | 20 |
| Total | | | 719 | | | 751 |

*Note:* m±SD – data is presented as mean and standard deviation; * – median (Interquartile range); ** – median (Minimum, Maximum). The results are rounded to thousandths


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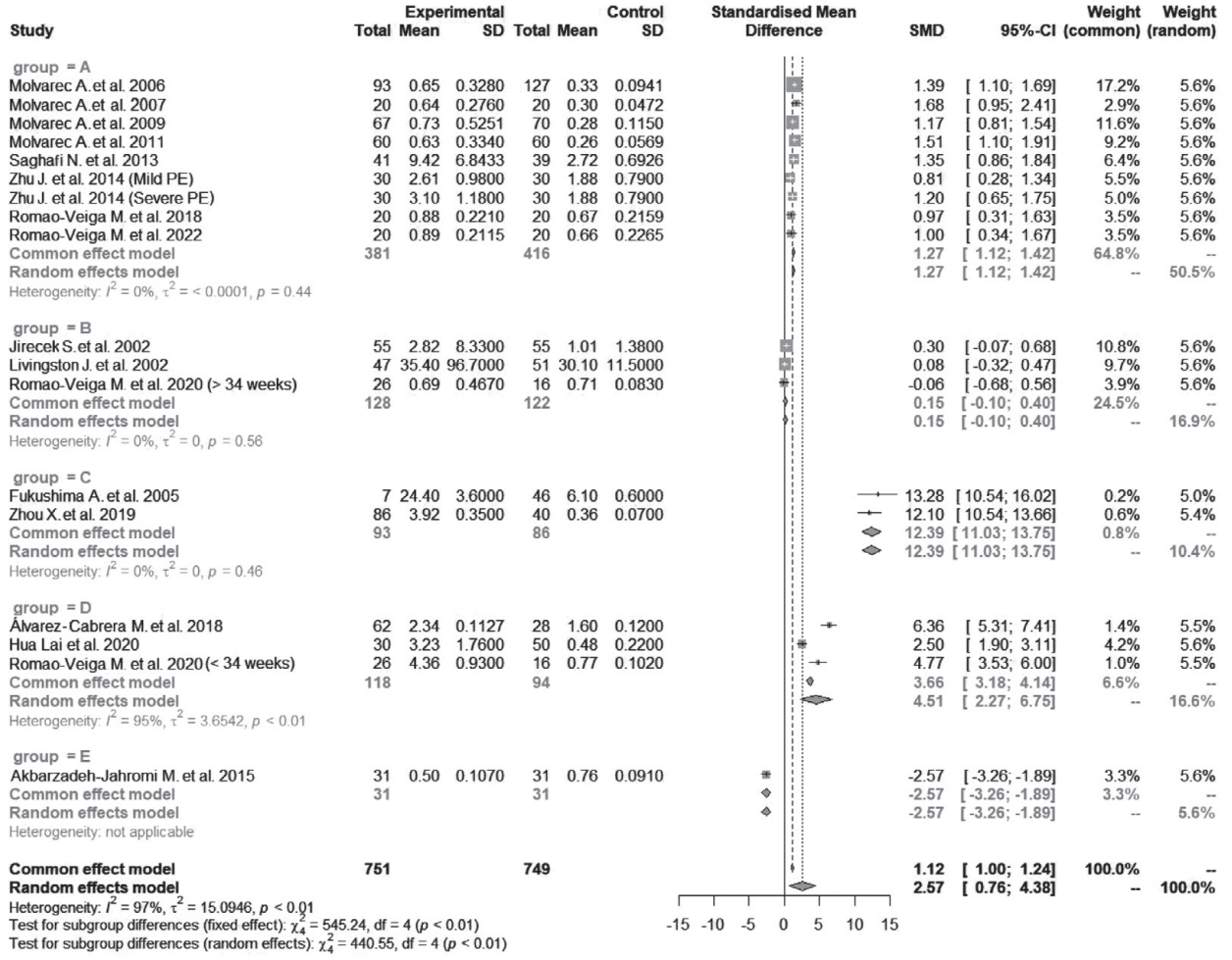


**Figure 2. Forest plot with division into groups**

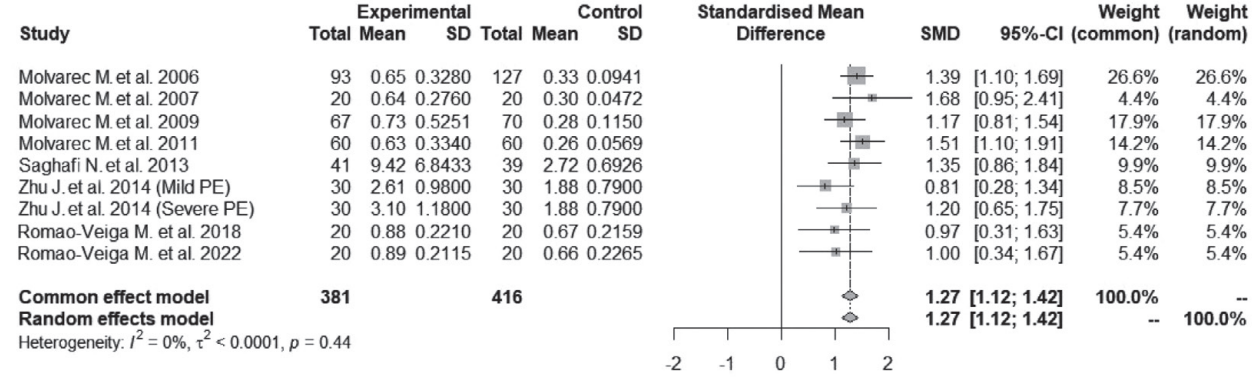


**Figure 2a. Forest plot of group A**

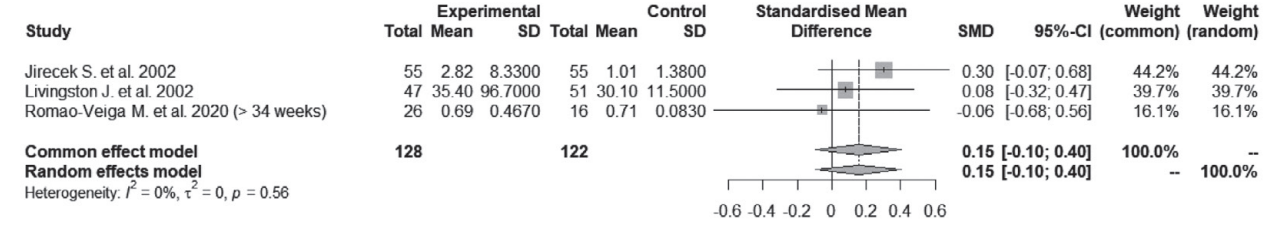


**Figure 2b. Forest plot of group B**



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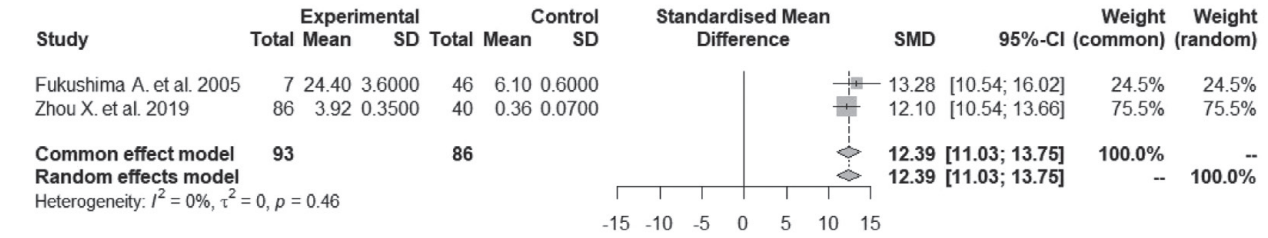


**Figure 2c. Forest plot of group C**

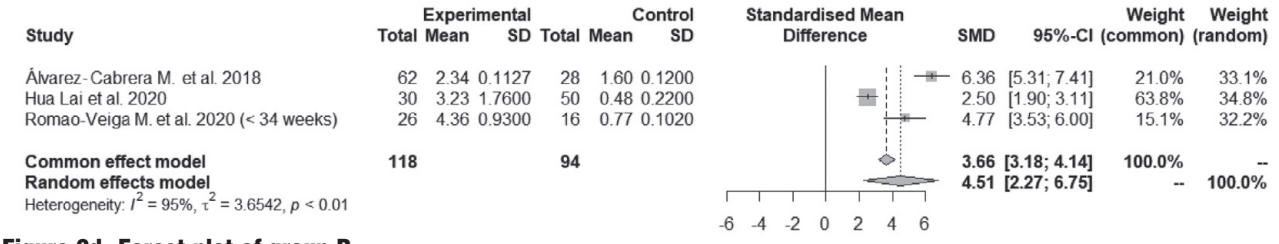


**Figure 2d. Forest plot of group D**

Out of 16 publications, only two (Romao-Veiga M. et al. 2020 [53] for group of gestational age less than 34 weeks, Akbarzadeh-Jahromi M. et al. 2015 [48]) had negative standardized difference means (worth noting, in Romao-Veiga M. et al. 2020 [53], the mean and standard deviation were estimated by the median and minimum and maximum values, which may cause errors).

The weighted mean SMD across all papers with the corresponding 95% confidence interval was 1.12 (1.00–1.24) for common-effects model and 2.57 (0.76–4.38) for random-effects model. Since both intervals do not contain 0, it can be assumed that the concentration of HSP70 in serum is significantly higher in patients with preeclampsia than in the control group at p<0.05.

The Cochrane Q-test showed high heterogeneity among studies (p<0,01), the value of the $I^2$ statistic was 97%. However, within most groups heterogeneity was lower: in group A (8 studies) $I^2$ was 0% (p=0.44), in group B (3 studies) – 0% (p=0.56), in group C (2 studies) – 0% (p=0.46), in group D (3 studies) – 95% (p<0.01). This high level of heterogeneity, but small within most groups, suggests certain factors are present that significantly influence some studies. The reason may be the lack of laboratory diagnostic standards and the use of diagnostic kits from different manufacturers.

The results of constructing meta-regression models are shown in Figure 3 ((a) is dependence of effect size SMD from maternal age, (b) – from gestational age, (c) – from systolic pressure (SP), (d) – from diastolic pressure (DP)). The corresponding coefficients «a», «b» of the linear regression $SMD=ax+b$, where x – parameter (maternal age, gestational age, SP and DP), are shown in Table 2.

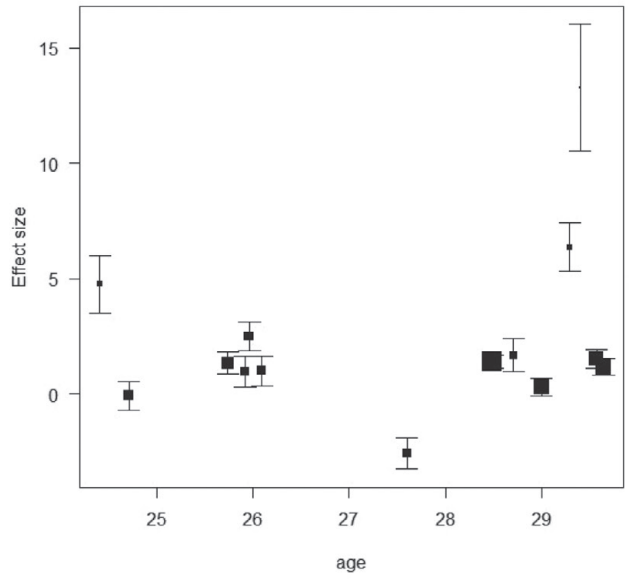


**Figure 3(a). Dependence of effect size SMD from maternal age**

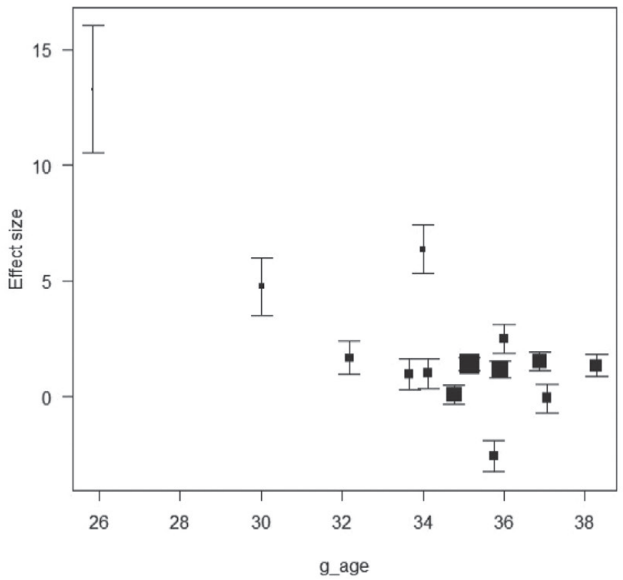


**Figure 3(b). Dependence of effect size SMD from gestational age**

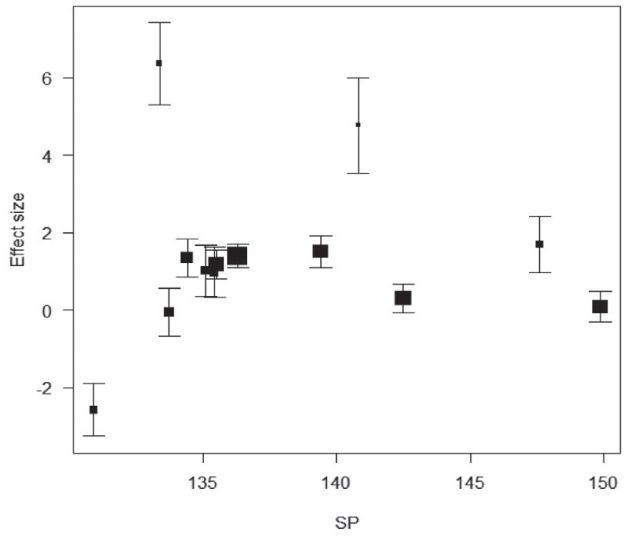


**Figure 3(c). Dependence of effect size SMD from SP**

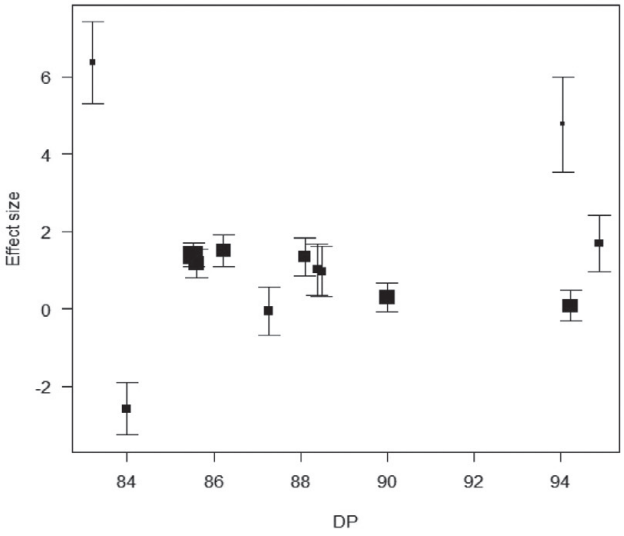


**Figure 3(d). Dependence of effect size SMD from DP**

*Table 2*

**Meta-regression coefficients**

| Model | a | *p* level | b | *p* level |
|---|---|---|---|---|
| Maternal age | 0.427 | 0.362 | -9.460 | 0.464 |
| Gestational age | -0.869 | <0.001 | 32.100 | <0.001 |
| Gestational age (excluded Fukushima A. et al. 2005) | -0.445 | 0.0689 | 17.100 | 0.0464 |
| Systolic pressure | 0.0156 | 0.879 | -0.808 | 0.955 |
| Diastolic pressure | 0.0273 | 0.859 | -1.06 | 0.937 |

Gestational age is the only parameter where coefficients «a», «b» differed from zero at a statistically significant level. However, if we remove the study of Fukushima A. et al. 2005 [43] (where SMD was significantly larger compared to others, and included only 7 cases of preeclampsia), the values cease to be statistically significant.

The funnel plot was built to test for publication bias in Figure 4(a). Most powerful studies (with standard error less than 0.4) fall within the 95% pseudo-confidence interval, being scattered symmetrically (Figure 4(a)). Egger's test accepted the hypothesis of publication bias at the level of p<0.01 (p=0.0155). However, it should be noted

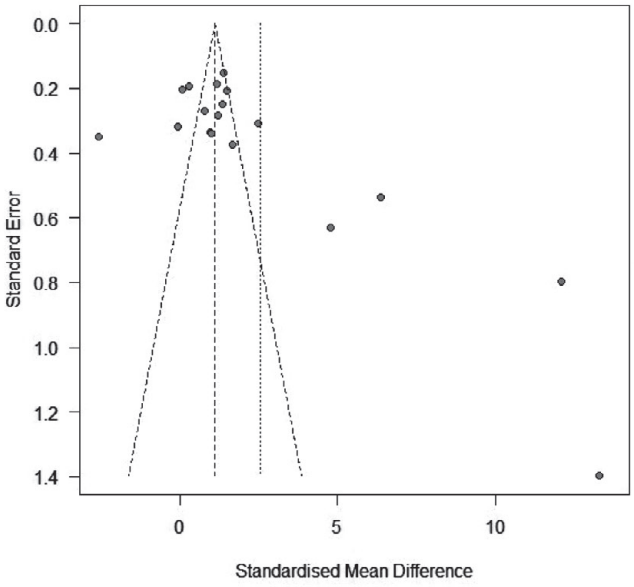


**Figure 4(a) Funnel-plot. Left dotted vertical line is weighted SMD from common-effects model, right – random-effects model**

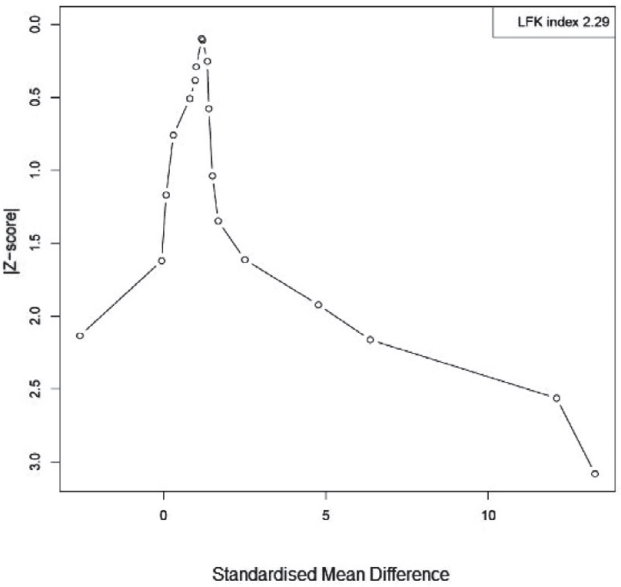


**Figure 4(b) DOI-plot. Each point corresponds publication, just as in funnel-plot, asymmetry is measured via LFK index**


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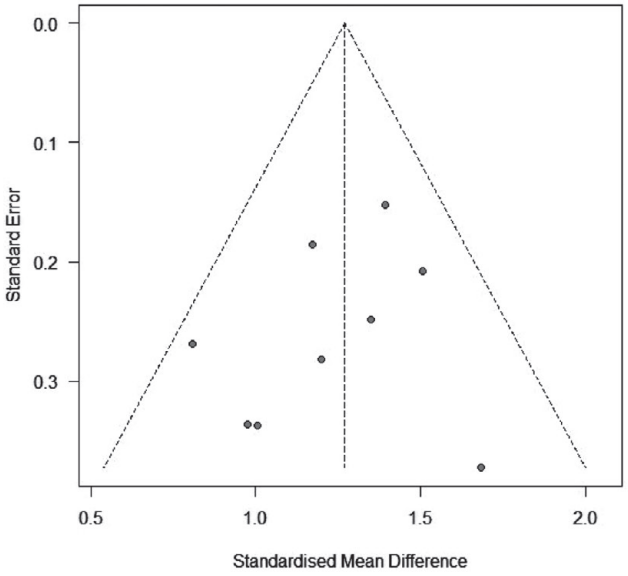



**Figure 5(a). Funnel plot for group A. Dotted line is weighted SMD from common-effects model**

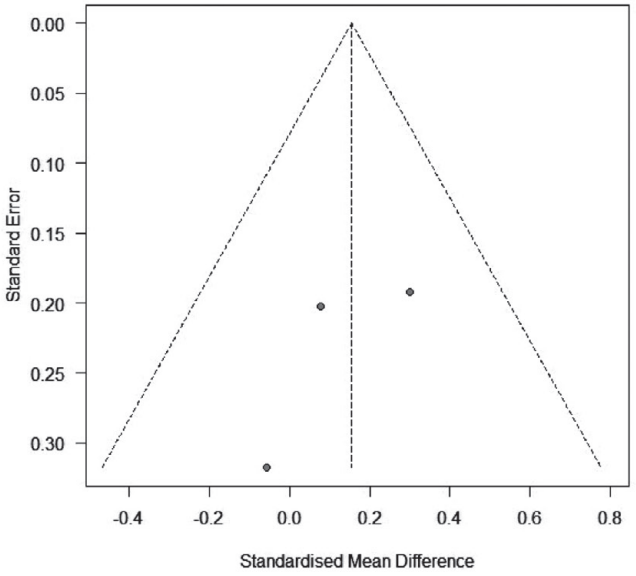



**Figure 5(b). Funnel plot for group B. Dotted line is weighted SMD from common-effects model**

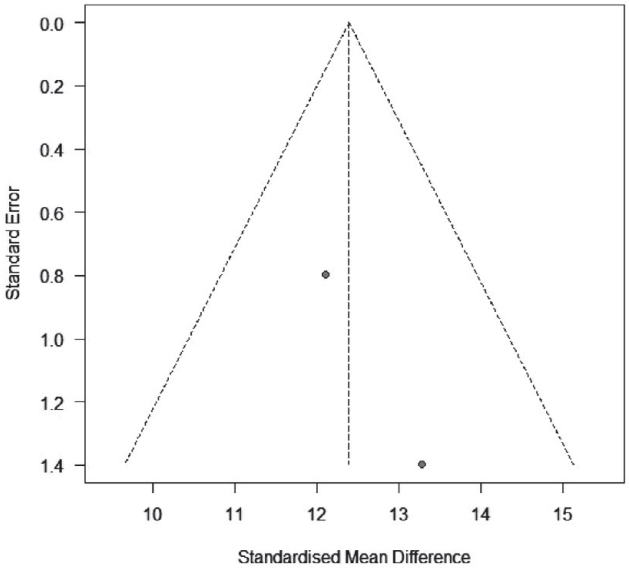



**Figure 5(c). Funnel plot for group C. Dotted line is weighted SMD from common-effects model**

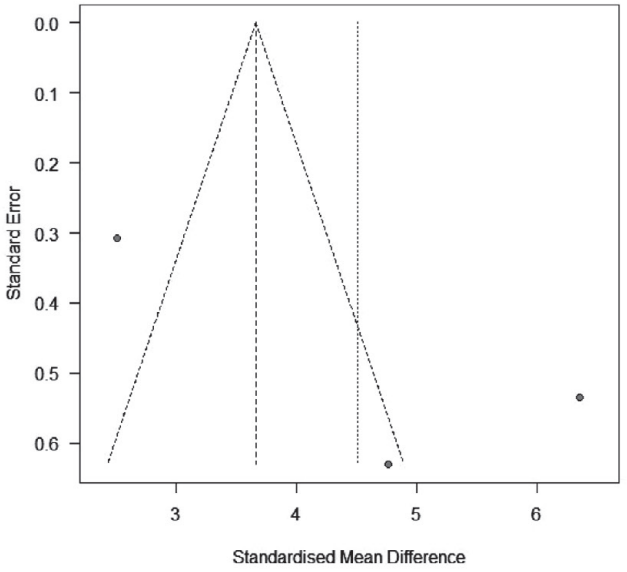



**Figure 5(d). Funnel plot for group D. Dotted line is weighted SMD from common-effects model**

that a lot of research is needed for sufficient power [39]. Hence, further research is necessary for confident conclusions. Figure 4(b) shows the DOI plot, the LFK index was 2.29, indicating significant asymmetry when considering all publications in general. This is one of the reasons why it is necessary to divide the works into groups and investigate whether the asymmetry was preserved.

For groups A, B, C, D Figure 5 ((a), (b), (c), (d) for each group) shows separate funnel plots using the common-effects model. It can be seen that all studies in groups A, B, C are inside the pyramid, scattered both in left and right part. Due to the small number of publications in each group, they were not checked by Egger's criterion.

DOI graphs for groups with LFK indices are shown in Figure 6 ((a), (b), (c), (d) for each group). For groups A, B, C, D, the indices were, respectively, -1.54, 2.53, 2.89, 3.39. So, there is moderate asymmetry in group A, and strong in groups B, C, and D. However, groups B, C, and D contained few elements, so drawing conclusions about asymmetry does not make sense. It is also worth remembering that for continuous variables there are cases when asymmetry in the funnel plot exists even in the absence of bias [55].

Sensitivity analysis via leave-one-out method gives reason to believe that the weighted estimate of the SMD value is stable and removing of one study (each in turn)

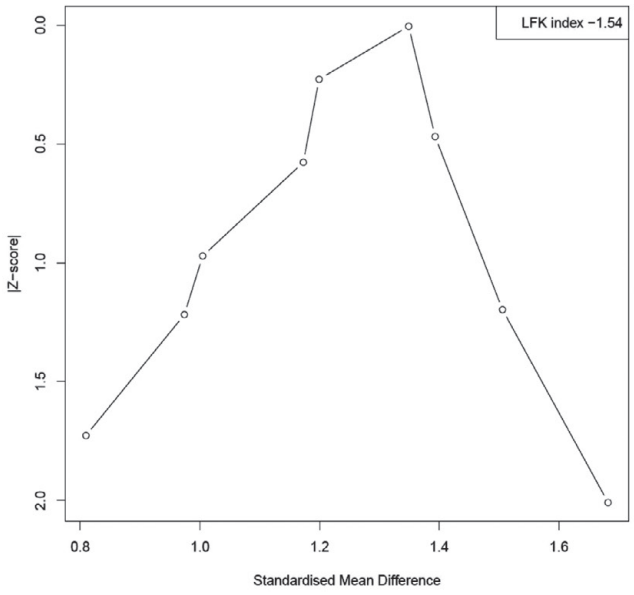
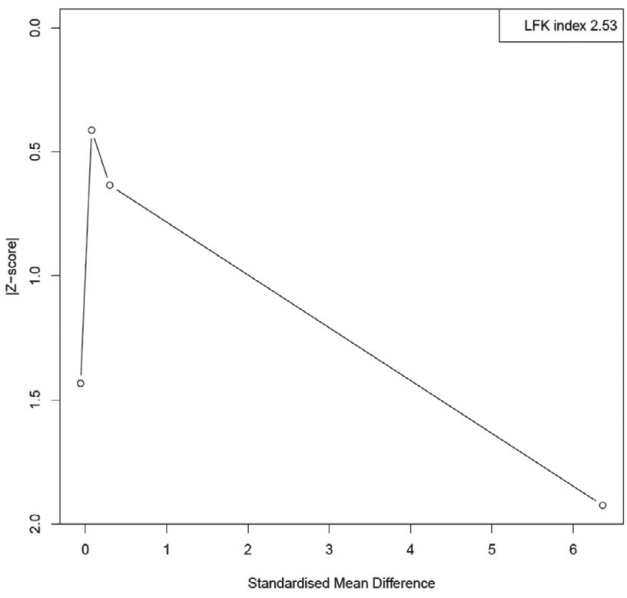



**Figure 6 (a). DOI plot for group A**




**A Figure 6 (b). DOI plot for group B**

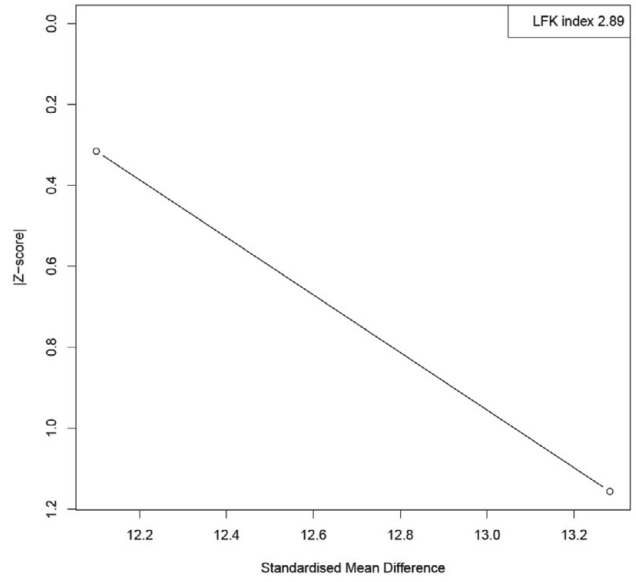



**Figure 6 (c). DOI plot for group C**

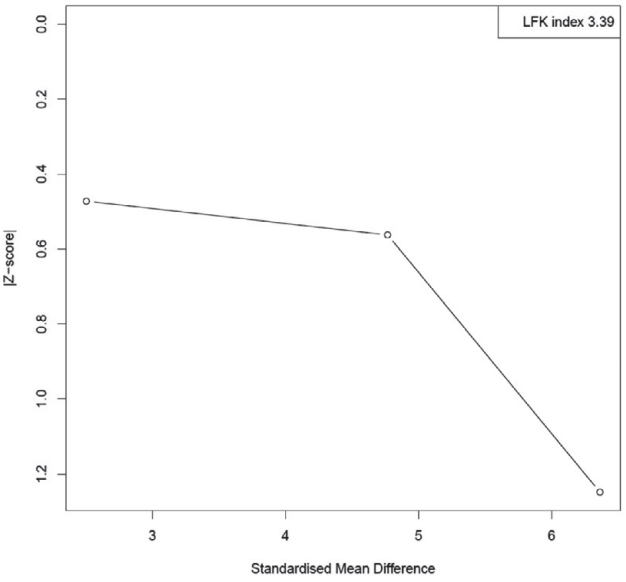



**Figure 6 (d). DOI plot for group D**

from the analysis does not change the statistical significance (Figure 7).

## DISCUSSION

There are 16 publications, which were included in the meta-analysis; in total, the study group included 751 pregnant women with preeclampsia and 719 healthy pregnant women as a control group. Works were identified via Scopus, PubMed Central, Virtual Health Library databases. One of meta-analysis limitations is that searches were not conducted in other regional databases (for example, Hindawi, National Library of China). Sometimes, lack of English translation of the article makes it impossible to be indexed by more well-known databases.

As for limitations of analysis, the absence of exact values of mean and standard deviation of HSP70 concentration in 8 studies could be mentioned. In 4 studies, they had to be estimated from median (range), and from median (interquartile range) in the other 4 using the Box-Cox transformation method. However, the error could be considered negligible. Insignificantly minor inaccuracy occurred from extracting data from plots using WebPlotDigitalizer in 2 studies.

The main limitation was the absence of laboratory diagnostics standards, use of diagnostic kits from differ-



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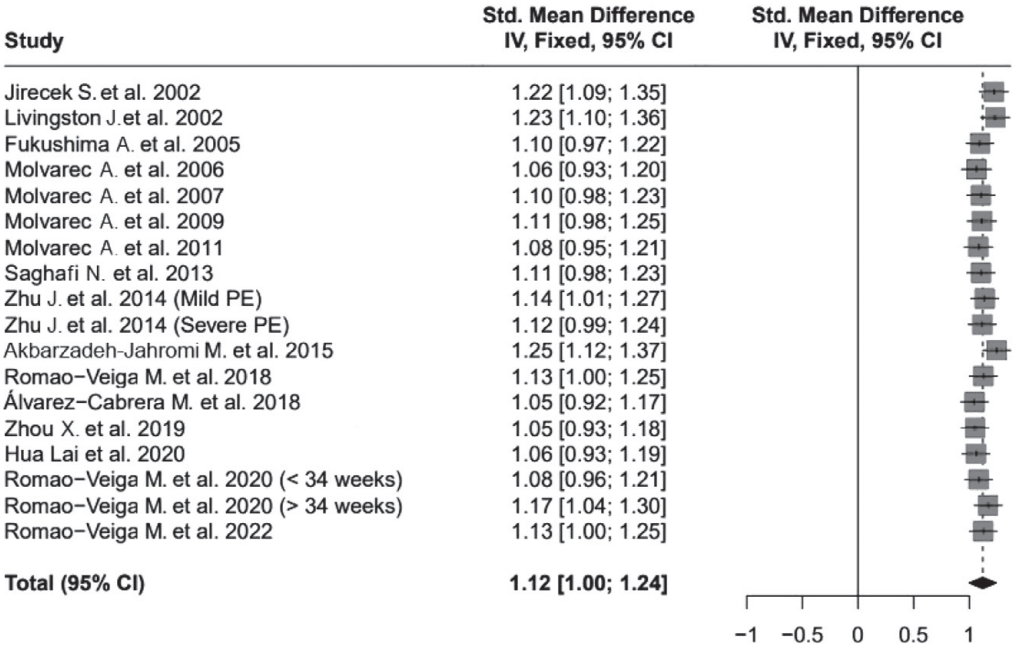


**Figure 7. Forest plot of sensitivity analysis in meta-analysis**

ent manufacturers by researchers, which eventually led to results that differed by 2 orders of magnitude. There is a study, where HSP70 serum concentration was measured with different enzyme-linked immunosorbent assay ELISA kits for every participant and then compared, the results varied significantly [56]. There were also differences in serum preparation (centrifugation for 5 or 10 minutes at 5000, 3000 or 1200 rpm) and in storage before the study (- 20 °C in two studies, - 70 °C in another one, - 80 °C in the remaining studies). The use of SMD analysis made it possible to compare the results obtained in different scales.

A limitation of all studies was that they were designed as case-control.

Some publications were excluded from the analysis. It is possible to examine work, published in 2013 by a group of researchers Jose C. Pera oli et al. (St. Paul, Brazil). The study included 237 pregnant women, divided by two groups with early preeclampsia onset and late one. Serum concentration of HSP60 and HSP70 were compared, their correlation with cytokine levels were determined [57]. HSP70 levels, obtained in group with early-onset preeclampsia, were significantly higher than those in women with late-onset preeclampsia. This study had a design limitation (did not report a control group of healthy pregnant women) and was therefore it was not included in meta-analysis.

In 2023, the research group Claudia M. Robellada-Zarate et al. (Mexico City, Mexico) addressed the topic of HSP70 during pregnancy [58]. It should be mentioned that this is the first prospective cohort study of HSP70. 48 pregnant women were included in the study, whose levels of HSP70, HSP60, and HSP27 were determined during the first screening examination at gestational age of 12 weeks. Researchers tried to find relationship between HSP level in the 1st trimester and preeclampsia development in the future. This publication was not included in the meta-analysis because it contained information on HSP70 in the first trimester.

There are also publications available, that report a change in HSP70 concentrations in blood serum or the expression of the corresponding genes in the placenta in pregnant women with the following obstetric complications of pregnancy: fetal growth restriction [33], fetoplacental hemodynamic disturbances [59], premature rupture of membranes and premature spontaneous labor [60] or chorioamnionitis [61]. When planning future studies of HSP70, these factors should be taken into account as potentially altering the picture and their presence or absence among study participants should be mentioned.

The studies included in the meta-analysis and reported serum HSP70 values in healthy and preeclamptic pregnant women, as well as maternal age and gestational age, body mass index (BMI), systolic and diastolic blood pressure at the time of examination, newborn weights are presented in Table 3. Not all publications contained data in full.

**Group A**

In terms of participants number, group A is the biggest group. Cochran's Q-test showed insignificant heterogeneity, $I^2$ was 0% (p=0.44), which indicates consistency of the obtained results. Four of the eight studies in group A were published by a group of researchers led by Attila Molvarec (Budapest, Hungary).

**A1.** In 2006, A. Molvarec et al. conducted a case-control study of HSP70 that included 142 pregnant women with hypertensive disorders during pregnancy and 127 normotensive pregnant women [5]. Of them, 93 had preeclampsia, 29 had gestational hypertension, and 20 had su-

*Table 3*

**Publications, included in meta-analysis, results of serum HSP70 measurements and other group characteristics**

| Authors | | Maternal age (years) | | Gestational age (weeks) | | BMI (kg/m²) | | Systolic blood pressure (mmHg) | | Diastolic blood pressure (mmHg) | | Fetal birth weight (kg) | | HSP70 Healthy | | HSP70 PE | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Healthy | PE | Healthy | PE | Healthy | PE | Healthy | PE | Healthy | PE | Healthy | PE | Median | n | Median | n |
| Jirecek S. et al. 2002 [41] | | 29±4.8 | 29±5.1 | - | - | - | - | 118 ± 21 | 167±21 | 75±14 | 105 ± 12 | 3.427± 0.62 | 2.985±0.814 | 1.01 ± 1.38 | 55 | 2.82 ± 8.33 | 55 |
| Livingston J. et al. 2002 [42] | | - | - | 35.2±6.0 | 34.3±5.6 | - | - | 127.0±23.4 | 174.7±17.2 | 79.7±13.8 | 110.0±12.1 | - | - | 30.1 ± 11.5 | 51 | 35.4 ± 96.7 | 47 |
| Fukushima A. et al. 2005 [43] | | 28.7±0.7 | 34.0±1.4 | 24.7±1.5 | 33.4±2.0 | - | - | - | - | - | - | - | - | 6.1 ± 0.6 | 46 | 24.4 ± 3.6 | 7 |
| Molvarec A. et al. 2006 [5] | | 28 (25-31) | 28 (25-32) | 35 (31-37) | 37 (35 - 39) | 26.0 (23.7-28.0) | 29.4 (26.3-32.0) | 110 (105-120) | 170 (160-180) | 70 (60-80) | 104 (100-115) | 3.3 (3.1-3.8) | 2.9 (1.98-3.45) | 0.31 (0.27-0.39)* | 127 | 0,55 (0.42 – 0.8)* | 93 |
| Molvarec A. et al. 2007 [44] | | 28.5 (26-32) | 27 (23-32.5) | 33 (30-35) | 33 (30-34) | 22 (19.7-24.2) | 21.5 (19.7-25.5) | 120 (110-120) | 180 (170-190) | 70 (70-80) | 120 (110-125) | 3.475 (3.1-3.65) | 1.665 (1.16-2.075) | 0.3 (0.27-0.33)* | 20 | 0.54 (0.47 – 0.79)* | 20 |
| Molvarec A. et al. 2009 [45] | | 30 (17-44) | 29 (19-42) | 35 (20-40) | 38 (30-41) | 25.9 (19.0-42.0) | 30 (20.6-38.3) | 110 (80-138) | 160 (135-220) | 70 (55-86) | 100 (90-131) | 3.5 (2.65-4.4) | 3.2 (1.4-4.2) | 0.28 (0.03-0.59)** | 70 | 0.58 (0.15-3.47)** | 67 |
| Molvarec A. et al. 2011 [46] | | 30 (28-32) | 29 (26-32) | 36 (36-37) | 37 (36-39) | 25.8 (24.3-27.9) | 29.9 (26.9-33.3) | 110 (107-120) | 162 (155-180) | 70 (60-80) | 100 (97-110) | 3.45 (3.15-3.7) | 3.125 (2.45-3.475) | 0.28 (0.21 – 0.31)* | 60 | 0.58 (0.39 – 0.81)* | 60 |
| Saghafi N. et al. 2013 [47] | | 26.3±5.6 | 25.2±4.9 | 38.8±2.3 | 37.8±5.2 | 24.1±0.4 | 25.6±3.5 | 113.9±10.5 | 153.9±16.2 | 69.6±8.3 | 105.7±10.8 | 3.083±0.541 | 2.722 ± 0.66 | 0 (0 - 4)* | 39 | 9 (0 – 23.5)* | 41 |
| Zhu J. et al. 2014 [34] | | - | - | - | - | - | - | - | - | - | - | - | - | 1.88 ± 0.79 | 30 | Mild PE<br>2.61 ± 0.98<br>Severe PE<br>3.10 ± 1.18 | 30<br><br><br>30 |
| Akbarzadeh-Jahromi M. et al. 2015 [48] | | 27.7±7.2 | 27.5±6.3 | 36.1±2.5 | 35.4±4.1 | - | - | 107.5±19.5 | 154.3±10.8 | 72.4±8.6 | 95.6±10.2 | - | - | 0.763 ± 0.091 | 31 | 0.504 ± 0.107 | 31 |
| Romao-Veiga M. et al. 2018 [49] | | 26 (14-41) | 25 (15-40) | 35 (24-39) | 34 (23-40) | - | - | 110 (90-112) | 160 (140-200) | 69 (63-70) | 110 (90-120) | - | - | 0.68023 (0.0075-1.0904)** | 20 | 0.9071 (0.4046-1.2728)** | 20 |
| Álvarez-Cabrera M. et al 2018 [50] | | 28.3±8.0 | 29.9±7.3 | 34.0±4.4 | 34.0±3.9 | 25.84±5.5 | 28.86±6.3 | 106.5±9.8 | 149.9±15.3 | 67.07±6.6 | 93.14±9.7 | - | - | 1.6 ± 0.12 | 28 | 2.338±0.1127 | 62 |
| Zhou X. et al. 2019 [0] | | - | - | - | - | - | - | - | - | - | - | - | - | 0.36 ± 0.07 | 40 | 3.92 ± 0.35 | 86 |
| Hua Lai et al. 2020 [52] | | 25.7±4.8 | 26.4±5.2 | 36 (36–37)* | 36 (36–38)* | 25.7±2.6 | 29.7±3.8 | - | - | - | - | - | - | 0.48 ± 0.22 | 50 | 3.23 ± 1.76 | 30 |
| Romao-Veiga M. et al. 2020 [53] | < 34 weeks | 23 (17-42) | 23 (14-41) | 30 (24-33) | 31 (23-33) | - | - | 105 (95-110) | 160 (140-210) | 65 (60-70) | 110 (90-140) | - | - | 0.8 (0.5 – 0.9)** | 16 | 4.5 (0.8 – 6.0)** | 26 |
| | > 34 weeks | 22 (15-39) | 24 (15-43) | 37 (34-40) | 37 (34-41) | - | - | 100 (90-110) | 150 (140-200) | 60 (60-70) | 100 (90-130) | - | - | 0.7 (0.6 – 0.9)** | 16 | 0.6 (0.09 – 1.9)** | 26 |
| Romao-Veiga M. et al. 2022 [54] | | 26 (17-39) | 25 (16-40) | 35 (27-39) | 34 (27-40) | - | - | 110 (90-112) | 160 (140-200) | 69 (63-70) | 110 (90-120) | - | - | 0.6735 (0.0075-1.0816)** | 20 | 0.9011 (0.4016-1.2638)** | 20 |

*Note:* PE – preeclampsia; m±SD – mean with standard deviation; * – median (Q1 – Q3). ** – median (min - max)

perimposed preeclampsia. A statistically significant difference was observed between HSP70 concentration in each group of women with hypertensive disorders compared to the control group. No statistically significant difference was found between serum HSP70 concentration in pregnant women with early and late-onset hypertensive disorders; as well as between groups of women with preeclampsia of varying severity; as well as between groups of gestational hypertension and superimposed preeclampsia. Also, no statistically significant differences of HSP70 levels were found between pregnant women with and without FGR, combined with hypertensive disorders. Thus, researchers concluded about inextricable link of HSP70 and pathogenesis of gestational hypertensive disorders. In our meta-analysis, from this research were included 93 pregnant with preeclampsia and 127 healthy pregnant women.

**A2.** In 2007, A. Molvarec et al. published a case-control study of HSP70 levels in 30 pregnant women with severe preeclampsia, 10 of whom also had evidence of HELLP syndrome [44]. The control group consisted of 20 pregnant women with a normal pregnancy. There was a statistically significant difference in HSP70 between pregnant women with severe preeclampsia and HELLP syndrome compared to healthy pregnant women. The authors hypothesized that HSP70 may be released into the bloodstream at sites of endothelial damage, as well as in the case of hepatocyte, platelet, and erythrocyte injury. In order to form a homogenous group 10 pregnant women with signs of HELLP were excluded from meta-analysis.

**A3.** A. Molvarec et al. in 2009 published the results of a study involving 67 preeclamptic women and 70 healthy pregnant women and found a relationship between HSP70 levels and markers of inflammation and oxidative stress, hepatocellular damage in the development of preeclampsia [45].

**A4.** In 2011, A. Molvarec et al. published another study, which included 60 pregnant women with preeclampsia and 60 pregnant women with uncomplicated pregnancies [4646]. Its aim was to investigate the relationship between HSP70 levels and levels of cytokines, chemokines, adhesion molecules and angiogenic factors in hypertensive complications of pregnancy. Elevated serum HSP70 concentrations in women with preeclampsia were associated with proinflammatory changes in the circulating cytokine profile. The authors hypothesized that circulating HSP70 may contribute to excessive systemic inflammatory response, specific to preeclampsia.

**A.5.** Group A also included the study of Jinming Zhu et al. from China, 2014, a survey in Xuzhou hospital, which included 90 pregnant women with hypertensive disorders during pregnancy (comprised of 30 with gestational hypertension, 30 with preeclampsia, 30 with preeclampsia with severe symptoms) and 30 healthy pregnant women [34]. HSP70 concentration in blood serum of pregnant women with preeclampsia and severe preeclampsia was found to be higher compared to groups of healthy pregnant women and women with gestational hypertension on statistically significant level ($p<0.05$). No statistically significant difference was found between levels of HSP70 in



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groups of gestational hypertension and healthy pregnant women (p>0.05). HSP70 concentration in the preeclampsia group with severe symptoms was significantly higher than in the preeclampsia group without severe symptoms (p<0.05). Thus, the researchers obtained data on the increase in HSP70 concentration depending on the severity of preeclampsia. 30 pregnant women with gestational hypertension were excluded from meta-analysis.

**A.6.** Researchers from Iran, Saghafi N. et al. 2013, studied HSP70 levels in 41 pregnant women with preeclampsia, 39 pregnant women in control group without extragenital and obstetric complications, and found a statistically significant difference in HSP70 between two groups [47]. It should be noted that serum was stored in this study at -20 °C.

**A.7.** Mariana Romao-Veiga et al. (Sao Paulo, Brazil), 2018, conducted a case-control study, which included 20 pregnant women with preeclampsia, 20 healthy pregnant women, and 20 nonpregnant women to study the association of preeclampsia with heat shock proteins and inflammatory markers [49]. The median of HSP70 concentration was higher in a group of pregnant women with preeclampsia compared to groups of women without preeclampsia and healthy non-pregnant women with a statistical significance (p<0.05).

**A.8.** Mariana Romao-Veiga et al. in 2022 continued a study of the relationship between preeclampsia and systemic inflammatory response [54]. Their case-control study included 20 pregnant women with preeclampsia and 20 healthy pregnant, 20 healthy non-pregnant women. The median of HSP70 concentration in the group of women with preeclampsia was higher on statistical significance p<0.05 compared to groups of healthy pregnant and healthy non-pregnant.

**Group B**

This group has three publications. In group B statistics $I^2$ was 0% (p=0.56), which suggest results are highly consistent.

**B.1.** In 2002, Stefan Jirecek (Austria) and colleagues studied HSP70 concentration in pregnant women with preeclampsia that occurred after 34 weeks (24 participants), pregnant women with preeclampsia that occurred before 34 weeks (31 participants); 55 normotensive pregnant women of corresponding gestational age were also included in the study [41]. Mean of HSP70 concentration in blood serum of women with early preeclampsia compared to women with late preeclampsia had a statistically significant difference (p<0.05). It should be noted that in this study, a large standard deviation compared to the mean must be interpreted as the presence of strong asymmetry to the right (i.e. there are a lot of HSP70 values that are much greater than defined mean). A statistically significant difference was obtained for HSP70 concentration in subgroups with early and late preeclampsia (p=0.01), thus it was concluded that HSP70 concentration was higher in pregnant women with early onset preeclampsia.

**B.2.** In 2002, US researchers Jeffrey C. Livingston and co-authors (USA, Tennessee Medical University) presented the results of their study, which investigated HSP70 concentration in 47 women with severe hypertension; the control group consisted of 51 normotensive pregnant women of corresponding gestational age [42]. No statistically significant difference in mean HSP70 levels in the two groups was found. In 28 pregnant women with severe preeclampsia and 30 controls, HSP70 concentration was below the sensitivity level of the kit, which was 0.0002 ng/ml. The researchers concluded that severe preeclampsia was not associated with increased serum HSP70 concentration. About material preparation for analysis: blood samples were centrifuged at 5000 rpm for 5 minutes; aliquots were stored at -70 °C until analysis.

**B3.** Research of Mariana Romao-Veiga et al. in 2020 was assigned to group B, although in 2018 and 2022 researchers used the same methods of laboratory research of HSP70 [54]. The reason was the design of study, which aimed to investigate early and late preeclampsia. High levels of HSP70 in women with early preeclampsia accounted for a different value of the standardized mean difference. 52 pregnant women with preeclampsia and 32 normotensive pregnant women were included. Both groups were divided in half: 26 pregnant with early and late gestosis each, 16 healthy pregnant women before and after 34 weeks. In early preeclampsia subgroup, the highest values of the median HSP70 was 4.5 (0.8 – 6.0) ng/ml (reported as median and IQR), which had a statistically significant difference to other subgroups (p<0.05). For late preeclampsia subgroup, the corresponding median and IQR for HSP70 was 0.6 (0.09–1.9) ng/ml, for controls subgroup up to 34 weeks 0.8 (0.5–0.9) ng/ml, for controls subgroup after 34 weeks it was 0.7 (0.6–0.9) ng/ml. This study did not report pooled results for preeclampsia group and healthy group. The difference of SMD in the subgroup of early and late preeclampsia was significant, therefore, the study of late preeclampsia was assigned to group B.

**Group C**

Group C includes two studies. Cochren's Q-test in group C resulted in $I^2$ about 0% (p=0.46), which suggests results consistency.

**C.1.** Researchers Xuru Zhou et al. from China (Hubei Province, Yichang City) in 2019 reported the results of a study aimed at finding a correlation between HSP70 and suppressor of cytokine signaling-3 SOCS-3 in pregnant women with preeclampsia [51]. 86 pregnant women with preeclampsia (including 35 pregnant women who had preeclampsia with severe symptoms), 40 healthy pregnant women made up the control group. The average HSP70 concentration in the preeclampsia group was different than in control group with a statistical significance (p<0.05). The risk of preeclampsia increased when HSP70 increased above 0.89 ng/ml, so the researchers considered it possible to use HSP70 as a marker of preeclampsia.

**C.2.** Researchers from Japan Akimune Fukushima et al. in 2005, studied HSP70 concentration in pregnant women at high risk of preterm birth (31 at risk of preterm birth and 7 with preeclampsia) and 46 healthy pregnant women [43]. No statistically significant difference in HSP70 levels during normal pregnancy between the three trimesters was found. Pregnant women with preeclamp-


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sia compared to women with normal pregnancies had a statistically significant difference (p=0.0001) in mean HSP70. The highest values of HSP70 were found in pregnant women who gave birth prematurely, the difference was statistically significant compared to healthy pregnant women (p=0.0005). Thus, according to the results of the study, higher values of HSP70 were found in pregnant women with preeclampsia and, especially, in treatment-resistant cases of premature birth. 7 preeclamptic women and 46 healthy pregnant women as the control group were included in the meta-analysis from this work.

**Group D**

Group D consists of three studies. Cochren's Q-test results in group D gave $I^2$ about 95% (p<0.01), which reflects high heterogeneity.

**D.1.** A group of researchers Hua Lai et al. from China (Jiangxi Province, Nanchang City) in 2020 published a study [52]. Apparently, it included the investigation of HSP70 in blood serum of 30 pregnant women with preeclampsia, 25 pregnant women with preeclampsia and FGR, and 50 healthy pregnant women as a control group. Levels of HSP70 in serum in three groups (healthy pregnant women, with preeclampsia, preeclampsia, and FGR) had a statistically significant difference (p<0.05).

**D.2.** Research by Mariana Romao-Veiga et al. in 2020 with early preeclampsia subgroup is assigned to group D [54]. No pooled results for preeclampsia and healthy groups were reported. Instead, results for early and late preeclampsia were reported, together with corresponding gestational age control groups. SMD differed in early and late preeclampsia groups significantly. Hence, the former was assigned to group D together with the corresponding control group.

**D.3.** María C. Álvarez-Cabrera (Mexico City, Mexico), 2018, presented a study that included 62 pregnant women with preeclampsia and 78 pregnant women as a control group (including 28 healthy pregnant women at 34 weeks and 50 healthy full-term pregnant women with onset childbirth) [50]. The median of HSP70 concentration in preeclampsia group was 1.5 times higher than the median HSP70 levels in healthy group at 34 weeks. Authors confidently concluded about the increase of HSP60 and HSP70 in pregnant women with preeclampsia compared to healthy pregnant women without labor activity. There was also a noticeable positive correlation of HSP60 and HSP70 with markers of the inflammatory response, and indicators characterizing liver dysfunction. From this study there were included 62 preeclamptic women and 28 healthy pregnant women without signs of labor.

**Group E**

In this group was put a study in which the levels of HSP70 in pregnant women with preeclampsia were compared to healthy pregnant women of corresponding age. In 2015, it was published by researchers Mojgan Akbarzadeh-Jahromi et al. from Iran, the city of Shiraz [48]. It included 2 groups: women with preeclampsia and healthy pregnant. In preeclampsia group, the average level of HSP70 was lower than in healthy group, but there was no statistically significant difference (p=0.310). Preservation conditions for serum before biochemical analysis were reported as -20 °C.

## CONCLUSIONS

The conducted meta-analysis makes it possible to confidently conclude about the increased in average HSP70 serum concentration in pregnant women with preeclampsia compared to healthy pregnant of the corresponding gestational age. No statistically significant relationship was found between increase of HSP70 concentration in preeclampsia and maternal age, gestational age, systolic and diastolic blood pressure. Data was insufficient to investigate via meta-regression models of association between HSP70 concentration in preeclampsia and parameters such as maternal body mass index and newborn weight.

Quantitative evaluation of HSP70 serum concentration is complicated by absence of single standard for laboratory diagnostics, which leads to difference in reported HSP70 values among different studies, sometimes by even 2 orders of magnitude. Study limitations was design type as case-control. The use of HSP70 as preeclampsia predictor is promising, but requires further study and conduct of prospective cohort studies.

The authors declare no conflict of interest regarding this paper.

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## REFERENCES


1. Say L, Chou D, Gemmill A, Tunçalp Ö, Moller AB, Daniels J, et al. Global causes of maternal death: a WHO systematic analysis. Lancet Glob Health. 2014;2(6):e323-33. doi: 10.1016/S2214-109X(14)70227-X.
2. Jung E, Romero R, Yeo L, Gomez-Lopez N, Chaemsaithong P, Jaovisidha A, et al. The etiology of preeclampsia. Am J Obstet Gynecol. 2022;226(2S):844-66. doi: 10.1016/j.ajog.2021.11.1356.
3. Jee B, Dhar R, Singh S, Karmakar S. Heat Shock Proteins and Their Role in Pregnancy: Redefining the Function of «Old Rum in a New Bottle». Front Cell Dev Biol. 2021;9:648463. doi: 10.3389/fcell.2021.648463.
4. Saghafi N, Pourali L, Ghavami GV, Mirzamarjani F, Mirteimouri M. Serum heat shock protein 70 in preeclampsia and normal pregnancy: A systematic review and meta-analysis. Int J Reprod Biomed. 2018;16(1):1-8.
5. Molvarec A, Prohászka Z, Nagy B, Szalay J, Füst G, Karádi I, et al. Association of elevated serum heat-shock protein 70 concentration with transient hypertension of pregnancy, preeclampsia and superimposed preeclampsia: a case-control study. J Hum Hypertens. 2006;20(10):780-6. doi: 10.1038/sj.jhh.1002060.
6. Brosens I, Pijnenborg R, Vercruysse L, Romero R. The «Great Obstetrical Syndromes» are associated with disorders of deep placentation. Am J Obstet Gynecol. 2011;204(3):193-201. doi: 10.1016/j.ajog.2010.08.009.
7. Rana S, Lemoine E, Granger JP, Karumanchi SA. Preeclampsia: Pathophysiology, Challenges, and Perspectives. Circ Res. 2019;124(7):1094-112. doi: 10.1161/CIRCRESAHA.118.313276. Erratum in: Circ Res. 2020;126(1):e8.
8. Popel OK, Govsieiev DO. Prospects of using sFlt-1, PlGF as biomarkers of superimposed preeclampsia. Ukr J Health of Woman. 2022;5(162):28-34. doi: 10.15574/HW.2022.162.28.
9. Portelli M, Baron B. Clinical Presentation of Preeclampsia and the Diagnostic Value of Proteins and Their Methylation Products as Biomarkers in Pregnant Women with Preeclampsia and Their Newborns. J Pregnancy. 2018;2018:2632637. doi: 10.1155/2018/2632637.
10. Tabacco S, Ambrosii S, Polsinelli V, Fantasia I, D'Alfonso A, Ludovisi M, et al. Pre-Eclampsia: From Etiology and Molecular Mechanisms to Clinical Tools-A Review of the Literature. Curr Issues Mol Biol. 2023;45(8):6202-15. doi: 10.3390/cimb45080391.
11. Anto EO, Ofori Boadu WI, Addai-Mensah O, Wiafe YA, Owiredu WK, Obirikorang C, et al. Association between micronutrients, oxidative stress biomarkers and angiogenic growth mediators in early and late-onset preeclamptic Ghanaian women. SAGE Open Med. 2023;11:20503121231175759. doi: 10.1177/20503121231175759.
12. Kampinga HH, Hageman J, Vos MJ, Kubota H, Tanguay RM, Bruford EA, et al. Guidelines for the nomenclature of the human heat shock proteins. Cell Stress Chaperones. 2009;14(1):105-11. doi: 10.1007/s12192-008-0068-7.
13. Brocchieri L, Conway de ME, Macario AJ. hsp70 genes in the human genome: Conservation and differentiation patterns predict a wide array of overlapping and specialized functions. BMC Evol Biol. 2008;8:19. doi: 10.1186/1471-2148-8-19.
14. Ellis RJ, Hemmingsen SM. Molecular chaperones: proteins essential for the biogenesis of some macromolecular structures. Trends Biochem Sci. 1989;14(8):339-42. doi: 10.1016/0968-0004(89)90168-0.
15. Bascos NAD, Landry SJ. A History of Molecular Chaperone Structures in the Protein Data Bank. Int J Mol Sci. 2019;20(24):6195. doi: 10.3390/ijms20246195.
16. Kurop MK, Huyen CM, Kelly JH, Blagg BSJ. The heat shock response and small molecule regulators. Eur J Med Chem. 2021;226:113846. doi: 10.1016/j.ejmech.2021.113846.
17. Dubrez L, Causse S, Borges Bonan N, Dumétier B, Garrido C. Heat-shock proteins: chaperoning DNA repair. Oncogene. 2020;39(3):516-29. doi: 10.1038/s41388-019-1016-y.
18. Rodriguez-Iturbe B, Lanaspa MA, Johnson RJ. The role of autoimmune reactivity induced by heat shock protein 70 in the pathogenesis of essential hypertension. Br J Pharmacol. 2019;176(12):1829-38. doi: 10.1111/bph.14334.
19. Rodríguez-Iturbe B, Johnson RJ. Heat shock proteins and cardiovascular disease. Physiol Int. 2018;105(1):19-37. doi: 10.1556/2060.105.2018.1.4.
20. Shan Q, Ma F, Wei J, Li H, Ma H, Sun P. Physiological Functions of Heat Shock Proteins. Curr Protein Pept Sci. 2020;21(8):751-60. doi: 10.2174/1389203720666191111113726.
21. Li DY, Liang S, Wen JH, Tang JX, Deng SL, Liu YX. Extracellular HSPs: The Potential Target for Human Disease Therapy. Molecules. 2022;27(7):2361. doi: 10.3390/molecules27072361.
22. Hu C, Yang J, Qi Z, Wu H, Wang B, Zou F, et al. Heat shock proteins: Biological functions, pathological roles, and therapeutic opportunities. MedComm. 2022;3(3):e161. doi: 10.1002/mco2.161.
23. Borges TJ, Murshid A, Theriault J, Calderwood SK. Molecular Chaperone Receptors: An Update. Methods Mol Biol. 2023;2693:193-208. doi: 10.1007/978-1-0716-3342-7_15.
24. Hagymasi AT, Dempsey JP, Srivastava PK. Heat-Shock Proteins. Curr Protoc. 2022;2(11):e592. doi: 10.1002/cpz1.592.
25. Bascos NAD, Landry SJ. A History of Molecular Chaperone Structures in the Protein Data Bank. Int J Mol Sci. 2019;20(24):6195. doi: 10.3390/ijms20246195.
26. Pockley AG. Heat shock proteins, inflammation, and cardiovascular disease. Circulation. 2002;105(8):1012-7. doi: 10.1161/hc0802.103729.
27. Álvarez-Cabrera MC, Barrientos-Galeana E, Barrera-García A, Osorio-Caballero M, Acevedo JF, Flores-Herrera O, et al. Secretion of heat shock -60, -70 kD protein, IL-1β and TNFα levels in serum of a term normal pregnancy and patients with pre-eclampsia development. J Cell Mol Med. 2018;22(11):5748-52. doi: 10.1111/jcmm.13824.
28. Rodriguez-Iturbe B, Johnson RJ, Sanchez-Lozada LG, Pons H. HSP70 and Primary Arterial Hypertension. Biomolecules. 2023;13(2):272. doi: 10.3390/biom13020272.
29. Tukaj S, Kaminski M. Heat shock proteins in the therapy of autoimmune diseases: too simple to be true? Cell Stress Chaperones. 2019;24(3):475-9. doi: 10.1007/s12192-019-01000-3.
30. Molvarec A, Rigo J Jr, Nagy B, Walentin S, Szalay J, Fust G, et al. Serum heat shock protein 70 levels are decreased in normal human pregnancy. J Reprod Immunol. 2007;74:163-9 doi:10.1016/j.jri.2006.12.002.
31. Tukaj S. Heat Shock Protein 70 as a Double Agent Acting Inside and Outside the Cell: Insights into Autoimmunity. Int J Mol Sci. 2020;21(15):5298. doi: 10.3390/ijms21155298.
32. Berestoviy VO, Mahmood A, Venckivska IB, Ginzburg VG, Sokol IV, Berestoviy OO, et al. The overview and role of heat shock proteins (HSP) especially HSP 60 and 70 in reproduction and other pathologies (a literature review) Medicni Perspektivi. 2021;26(1):54-62. doi: 10.26641/2307-0404.2021.1.227733.
33. Lai H, Nie L, Zeng X, Xin S, Wu M, Yang B, et al. Enhancement of heat shock protein 70 attenuates inducible nitric oxide synthase in preeclampsia complicated with fetal growth restriction. J Matern Fetal Neonatal Med. 2022;35(13):2555-63. doi: 10.1080/14767058.2020.1789965.
34. Zhu J, Li M, Li L. Expression and significance of heat shock protein 70 in maternal serum, umbilical cord blood and placenta of patients with hypertensive disorders complicating pregnancy. Zhonghua Fu Chan Ke Za Zhi. 2014;49(9):676-80.
35. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. BMJ. 2021;372:n71. doi: 10.1136/bmj.n71.
36. McGrath S, Zhao X, Steele R, Thombs BD, Benedetti A; DEPRESsion Screening Data (DEPRESSD) Collaboration. Estimating the sample mean and standard deviation from commonly reported quantiles in meta-analysis. Stat Methods Med Res. 2020;29(9):2520-37. doi: 10.1177/0962280219889080.
37. McGrath S, Zhao X, Steele R, Benedetti A. Estmeansd: Estimating the Sample Mean and Standard Deviation from Commonly Reported Quantiles in Meta-Analysis_R package version 1.0.0 [Internet]. 2022. Available from: https://CRAN.R-project.org/package=estmeansd.
38. Higgins JP, Thomas J, Chandler J, Cumpston M, Li T, Page MJ, editors. Cochrane Handbook for Systematic Reviews of Interventions, 2nd Edition [Internet]. New York: Wiley-Blackwell; 2019. 736 p. Available from: https://training.cochrane.org/handbook.
39. Sterne J, Egger M. Regression Methods to Detect Publication and Other Bias in Meta-Analysis. Publication Bias in Meta-Analysis: Prevention, assessment and adjustments. 2006:99-110. doi: 10.1002/0470870168.ch6.
40. Furuya-Kanamori L, Barendregt JJ, Doi SAR. A new improved graphical and quantitative method for detecting bias in meta-analysis. Int J Evid Based Healthc. 2018;16(4):195-203. doi: 10.1097/XEB.0000000000000141.
41. Jirecek S, Hohlagschwandtner M, Tempfer C, Knöfler M, Husslein P, Zeisler H. Serum levels of heat shock protein 70 in patients with preeclampsia: a pilot-study. Wien Klin Wochenschr. 2002;114(15-16):730-2.
42. Livingston JC, Ahokas R, Haddad B, Sibai BM, Awaads R. Heat shock protein 70 is not increased in women with severe preeclampsia. Hypertens Pregnancy. 2002;21(2):123-6. doi: 10.1081/PRG-120004767.
43. Fukushima A, Kawahara H, Isurugi C, Syoji T, Oyama R, Sugiyama T, et al. Changes in serum levels of heat shock protein 70 in preterm delivery and preeclampsia. J Obstet Gynaecol Res. 2005;31(1):72-7. doi: 10.1111/j.1447-0756.2005.00244.x.
44. Molvarec A, Prohászka Z, Nagy B, Kalabay L, Szalay J, Füst G, et al. Association of increased serum heat shock protein 70 and C-reactive protein concentrations and decreased serum alpha(2)-HS glycoprotein concentration with the syndrome of hemolysis, elevated liver enzymes, and low platelet count. J Reprod Immunol. 2007;73(2):172-9. doi: 10.1016/j.jri.2006.07.002.
45. Molvarec A, Rigó J Jr, Lázár L, Balogh K, Makó V, Cervenak L, et al. Increased serum heat-shock protein 70 levels reflect systemic inflammation, oxidative stress and hepatocellular injury in preeclampsia. Cell Stress Chaperones. 2009;14(2):151-9. doi: 10.1007/s12192-008-0067-8.

46. Molvarec A, Szarka A, Walentin S, Beko G, Karádi I, Prohászka Z, et al. Serum heat shock protein 70 levels in relation to circulating cytokines, chemokines, adhesion molecules and angiogenic factors in women with preeclampsia. Clin Chim Acta. 2011;412(21-22):1957-62. doi: 10.1016/j.cca.2011.06.042.
47. Saghafi N, Hoseini HA, Amel JS, Ghazanfari SM, Namani H. Comparison of Serum Heat-Shock Protein 70 Levels in Pre-Eclampsia with Normal Pregnancy. Iranian J Obst, Gynecol Infertility. 2013;16(70):1-8. doi: 10.22038/ijogi.2013.1925.
48. Akbarzadeh-Jahromi M, Daneshyar Z,A Slani FS, Asadi N, Zare HR. Circulating levels of heat shock protein 70 in women with preeclampsia and healthy controls. Shiraz E Med J. 2015;16(6): e27750.
49. Romão-Veiga M, Matias ML, Ribeiro VR, Nunes PR, M Borges VT, Peraçoli JC, et al. Induction of systemic inflammation by hyaluronan and hsp70 in women with pre-eclampsia. Cytokine. 2018;105:23-31. doi: 10.1016/j.cyto.2018.02.007.
50. Álvarez-Cabrera MC, Barrientos-Galeana E, Barrera-García A, Osorio-Caballero M, Acevedo JF, Flores-Herrera O, et al. Secretion of heat shock -60, -70 kD protein, IL-1β and TNFα levels in serum of a term normal pregnancy and patients with pre-eclampsia development. J Cell Mol Med. 2018;22(11):5748-52. doi: 10.1111/jcmm.13824.
51. Xuru ZHOU, Lingling XIE, Hongyu XU, Xi WANG, Jie TIAN, Xiaolan LI Relationship among serum heat shock protein 70, suppressor of cytokine signaling-3 and immune factor in pregnant women with hypertension and its diagnostic value. Clin Med China. 2019;(12):63-8.
52. Lai H, Nie L, Zeng X, Xin S, Wu M, Yang B, et al. Enhancement of heat shock protein 70 attenuates inducible nitric oxide synthase in preeclampsia complicated with fetal growth restriction. J Matern Fetal Neonatal Med. 2022;35(13):2555-63. doi: 10.1080/14767058.2020.1789965.
53. Romão-Veiga M, Bannwart-Castro CF, Borges VTM, Golim MA, Peraçoli JC, Peraçoli MTS. Increased TLR4 pathway activation and cytokine imbalance led to lipopolysaccharide tolerance in monocytes from preeclamptic women. Pregnancy Hypertens. 2020;21:159-65. doi: 10.1016/j.preghy.2020.06.002.
54. Romao-Veiga M, Ribeiro VR, Matias ML, Nunes PR, Romagnoli GG, Peracoli JC, et al. DAMPs are able to skew CD4+ T cell subsets and increase the inflammatory profile in pregnant women with preeclampsia. J Reprod Immunol. 2022;149:103470. doi: 10.1016/j.jri.2021.103470.
55. Freeman S, Sutton A. Identifying publication bias in meta-analyses of continuous outcomes. 2020. Webinar. Available from: https://training.cochrane.org/resource/identifying-publication-bias-meta-analyses-continuous-outcomes.
56. Njemini R, Demanet C, Mets T. Comparison of two ELISAs for the determination of Hsp70 in serum. J Immunol Methods. 2005;306(1-2):176-82. doi: 10.1016/j.jim.2005.08.012.
57. Peraçoli JC, Bannwart-Castro CF, Romao M, Weel IC, Ribeiro VR, Borges VT, et al. High levels of heat shock protein 70 are associated with pro-inflammatory cytokines and may differentiate early-from late-onset preeclampsia. J Reprod Immunol. 2013;100(2):129-34. doi: 10.1016/j.jri.2013.08.003.
58. Robellada-Zárate CM, Luna-Palacios JE, Caballero CAZ, Acuña-González JP, Lara-Pereyra I, González-Azpeitia DI, et al. First-trimester plasma extracellular heat shock proteins levels and risk of preeclampsia. J Cell Mol Med. 2023;27(9):1206-13. doi: 10.1111/jcmm.17674.
59. Kao CK, Morton JS, Quon AL, Reyes LM, Lopez-Jaramillo P, Davidge ST. Mechanism of vascular dysfunction due to circulating factors in women with pre-eclampsia. Clin Sci (Lond). 2016;130(7):539-49. doi: 10.1042/CS20150678.
60. Dvorakova L, Ivankova K, Krofta L, Hromadnikova I. Expression profile of heat shock proteins in placental tissues of patients with preterm prelabor rupture of membranes and spontaneous preterm labor with intact membranes. Am J Reprod Immunol. 2017;78(4). doi: 10.1111/aji.12698.
61. Osorio-Caballero M, Perdigón-Palacio C, García-López G, Flores-Herrera O, Olvera-Sánchez S, Morales-Méndez I, et al. Escherichia coli-induced temporal and differential secretion of heat-shock protein 70 and interleukin-1β by human fetal membranes in a two-compartment culture system. Placenta. 2015;36(3):262-9. doi: 10.1016/j.placenta.2014.12.011.

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