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Activity of antioxidant enzymes in different pathophysiological states

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Abstract

Background: Oxidative stress is supposed to be implicated in the pathogenesis of several diseases which are connected with increased formation of reactive oxygen and nitrogen species (RONS). Organisms are protected against RONS from antioxidant system that is composed of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase, glutathione reductase and paraoxonase (PON) and non-enzymatic antioxidants. The aim of this doctoral thesis was to investigate the behaviour of three of these antioxidant enzymes ï CuZnSOD, CAT and PON1 in different pathophysiological states.

Materials and methods: The activities of CuZnSOD, CAT and PON1 were measured in 40 patients with metabolic syndrome (MetS), 35 women with depressive disorder (DD), 30 septic patients (SP), 50 patients with pancreatic cancer (PC), 50 patients with chronic pancreatitis (CP) and 13 patients with acute pancreatitis (AP) together with sex- and age-matched healthy controls (CON). In all these studies also the levels of oxidative stress markers were measured.

Results: The activity of CAT was found to be decreased in patients with sepsis or septic shock, MetS and PC in comparison with CON, while in patients with DD, CP and AP no differences in CAT activity were detected. The activities of CuZnSOD were in the contrast to CAT either increased or unaffected. Increased activities of CuZnSOD were observed in MetS, DD, PC and SP, while no differences in CuZnSOD activities were found between CP or AP and CON. In all observed pathophysiological states the arylesterase activity of PON1 was measured and was found to be decreased (with the exception of DD) in comparison with CON.

Conclusion: It was shown, that all selected diseases are connected with increased oxidative stress, which leads to the changes in antioxidant enzymes activities.

Abstrakt

Úvod: U onemocnění spojených se zvýšenou tvorbou reaktivních sloučenin kyslíku a dusíku (RONS) se předpokládá účast oxidačního stresu v jejich patogenezi. Organismus je proti působení RONS chráněn antioxidačním systémem, který je tvořen antioxidačními enzymy jako superoxiddismutasa (SOD), katalasa (CAT), glutathionperoxidasa (GPX), glutathionreduktasa (GR) či paraoxonasa (PON) a neenzymatickými antioxidanty. Cílem této disertační práce bylo vyšetření aktivity tří těchto antioxidačních enzymů – SOD, CAT, PON za různých patofyziologických stavů.

Materiál a metody: Aktivity CuZnSOD, CAT a PON1 byly měřeny u 40 pacientů s metabolickým syndromem (MetS), 35 žen s depresivní poruchou (DD), 30 pacientů se sepsí (SP), 50 pacientů s karcinomem pankreatu (PC), 50 pacientů s chronickou pankreatitidou (CP) a 13 pacientů s akutní pankreatitidou (AP) a ke každé sledované skupině pacientů byla zařazena též kontrolní skupina spárovaná na základě věku a pohlaví (CON). Ve všech studiích byly zároveň měřeny markery oxidačního stresu.

Výsledky: Snížené hladiny aktivit CAT byly pozorovány u pacientů se sepsí či septickým šokem, MetS a PC v porovnání s kontrolami, zatímco u pacientů s DD, CP a AP nebyly zjištěny žádné rozdíly v aktivitě CAT při srovnání s CON. U pacientů s MetS, DD, PC a SP byly zjištěny zvýšené aktivity CuZnSOD, i když u pacientů s CP a AP nebyly pozorovány rozdíly v aktivitě CuZnSOD při srovnání s CON. U všech sledovaných patofyziologických stavů (s výjimkou depresivních poruch) byly nalezeny snížené aktivity PON1 v porovnání s CON.

Závěr: Provedené studie ukazují, že všechna sledovaná onemocnění jsou spojena se zvýšeným oxidačním stresem, v rámci kterého dochází k ovlivnění chování námi sledovaných antioxidačních enzymů.

1. Introduction

Antioxidants are compounds that control the redox-balance in biological systems. Even in small concentrations they prevent the oxidation of instable substrates and/or eliminate the reactive oxygen and nitrogen species (RONS). Antioxidants represent heterogeneous group of chemical compounds., They could be divided according to their character to enzymatic and non-enzymatic antioxidants. To the most important antioxidant enzymes belong superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPX), glutathione reductase (GR), and paraoxonase (PON). The most important non-enzymatic antioxidant is reduced glutathione (GSH).

In physiological conditions there is equilibrium between RONSôs formation and their degradation in organism due to the antioxidants. Overproduction of RONS in connection with impaired function of antioxidant system leads to oxidative stress. It was shown that RONS and/or oxidative stress could be involved in the pathogenesis of some diseases such as atherosclerosis, diabetes mellitus, inflammation and sepsis or neurodegenerative diseases. The role of RONS in clinical medicine was in detail described by Macasek et al. (2011). The research of RONS and antioxidants became actual theme in medicine in the recent years.

2. Aims and scopes

The aim of this doctoral thesis was to investigate the behaviour of three antioxidant enzymes ï superoxide dismutase, catalase, and paraoxonase in different pathophysiological states.

This thesis was focused on antioxidant enzymes activities and their changes in acute phase of the disease such as sepsis or acute pancreatitis, where the activities were followed up in the course of sepsis or acute pancreatitis.

In chronic states such as metabolic syndrome, pancreatic carcinoma or chronic pancreatitis the antioxidant enzymes activities were compared with those of healthy volunteers.

3. Materials and methods

Patients:

The activities of antioxidant enzymes (CAT, CuZnSOD and PON1) and levels oxidative stress markers such as conjugated dienes in precipitated LDL (CD-LDL) or oxidized-LDL (ox-LDL/LDL) were measured in patients with different diseases. The activities were investigated in group of 40 patients with metabolic syndrome, of 35 depressive women, of 30 septic patients, of 45 patients with chronic pancreatitis 45 patients with carcinoma of pancreas and 13 patients with acute pancreatitis.

All groups of patients were compared with sex- and age- matched healthy controls.

Blood sample:

Blood samples were obtained after overnight fasting. Activities of CAT and CuZnSOD were measured in haemolysed erythrocytes. The blood samples were collected into the tubes with K_2EDTA, erythrocytes were washed three times with a NaCl isotonic solution (9g/l). Serum was used for the determination of PON1. The samples were stored at -80°C until assay.

Enzymes activities:

Catalase: The activity was determined by the modified method of Aebi. The reaction mixture in cuvettes contained 876 μl of 50 mM potassium phosphate buffer, pH = 7.2 and 25 μl of diluted sample. The reaction was started after 10 minutes of incubation at 30 ºC by addition of 99 μl of 10 mM H_2O_2. The rate of H_2O_2 degradation was monitored spectrophotometrically at 240 nm. Catalase activity was calculated using the molar extinction coefficientof $H_2O_2$43.6 M^{-1} cm^{-1}.

CuZn-Superoxide dismutase: The activity was determined according to the modified method of Ġtípek et al. The reaction mixture in cuvettes contained 700 µl of 50 mM potassium phosphate buffer, pH = 7.2; 50 µl of xanthine oxidase; 100µl of NBT and 50 µl of diluted sample. The reaction was started after 10 minutes of incubation at 25 ºC by addition of 100 µl of 1 mM xanthine. The rate of NBT-formazan generation was monitored spectrophotometrically at 540 nm. Superoxide dismutase activity was calculated by means of calibration curve.

Paraoxonase 1: The arylesterase activity of PON1 was measured according to the method of Eckerson et al. using phenylacetate as a substrate. Briefly, 900 µl of 20mMTris-HCl buffer containing 1 mM $CaCl_2$, pH=8.0 was added to cuvettes followed by 50 µl of diluted serum sample. The reaction was started by addition of 50µl of 100 mM phenylacetate. The rate of phenol generation was monitored spectrophotometrically at 270 nm. Arylesterase activity of PON1 was calculated using the molar extinction coefficient of the produced phenol, 1310 $M^{-1}cm^{-1}$.

Paraoxonase activity of PON1 was measured using paraoxon (O,O-Diethyl O-(4-nitrophenyl) phosphate) as a substrate in tubes containing 940 µl of 90 mM Tris-buffer (pH = 8.5, with 2 mM $CaCl_2$) and 50 µl of 100 mM paraoxon. The reaction was started by addition of 10 µl of serum and measured at 405 nm at 25°C. The activity was calculated using the molar extinction coefficient of the produced p-nitro-phenol, 18053 $M^{-1}cm^{-1}$.

The lactonase activity of PON1 was determined according to the modified method of Draganov et al. (2000). Briefly, to 800 µl of 50 mM TRIS-buffer (pH = 8, with 1mM $CaCl_2$) in cuvettes 100 µl of diluted serum sample was added. The reaction was started after incubation at 30°C for 5 minutes with 100 µl of dihydrocoumarin (final concentration 1mM). The increase in absorbance at 270 nm was monitored along 2 minutes. Kinetic rate was estimated during the linear phase of reaction

and converted to enzyme activity using molar extinct coefficient of the reaction product 3-(2-hydroxy-phenyl)-propionate ($\mathring{U}$= 1295 $dm^3mol^{-1}cm^{-1}$), (Rainwater, 2009).

All statistical analyses were performed using version 8.0 of StatSoft software Statistica (2007, CZ).

4. Results

Activities of three different antioxidant enzymes in erythrocytes: CuZnSOD, CAT and PON1 together with markers of oxidative stress in serum ï CD-LDL, ox-LDL and NT were established in six different pathophysiological states. Furthermore serum concentrations of enzymatic cofactors such as Cu, Zn, Fe and Ca and levels of some lipid parameters ï HDL-C and apo-A1 and in some studies concentrations of SAA were determined. Patients with metabolic syndrome, depressive disorder, sepsis or septic shock, pancreatic carcinoma, chronic and acute pancreatitis were included into the studies dealing with oxidative stress and status of antioxidant defence system.

In all these above mentioned diseases increased levels of oxidative stress markers were observed compared to CON.

The results of antioxidant enzymes activities in different pathophysiological states are summarized in the Table 1. The activity of CAT was found to be decreased in patients with sepsis or septic shock, MetS and PC in comparison with CON, while in patients with DD, CP and AP no differences in CAT activity were detected. The activities of CuZnSOD were in the contrast to CAT either increased or unaffected. Increased activities of CuZnSOD were observed in MetS, DD, PC and SP, while no differences in CuZnSOD activities were found between CP or AP and CON. In all observed pathophysiological states the arylesterase activity of PON1 was measured and was found to be decreased (with the exception of DD) in comparison with CON.

	CAT(kU/g Hb)	CuZnSOD(kU/g Hb)	PON1(U/ml)
Metabolic syndrome (n = 40, M/F = 20/20)	189.6 ± 31.8*	2.3 (1.9 - 2.5)**	158.9 ± 41.9*
Controls (n = 40, M/F = 20/20)	204.6 ± 33.0	2.0 (1.2 - 2.5)	179.9 ± 42.3
Depressive disorder (n = 35, F)	174.0 (155.2 ï 217.2)	2.4 (2.1 - 2.6)***	161.3 (140.8 - 196.2)
Controls (n = 35, F)	189.0 (166.6 ï 215.4)	1.9 (1.3 ï 2.2)	175.9 (146.2 ï 207.3)
Septic patients (n = 30, M/F = 12/18)	169.2 ± 32.0*	1.9 ± 0.5***	89.5 ± 43.2***
Controls (n = 30, M/F = 12/18)	198.3 ± 32.7	1.2 ± 0.2	172.0 ± 48.7
Carcinoma of pancreas (n = 45, M/F = 28/17)	182.6 (168.3-204.2)***$^{+}$	2.5 (2.3 - 2.6)$^{++}$	114.8 (88.7-131.4)***$^{+}$
Chronic pancreatitis (n = 45, M/F = 28/17)	197.2 (177.1-232.0)	1.7 (1.5 ï 2.0)	124.4 (104.7-151.7)**
Controls (n = 45, M/F = 28/17)	205.1 (179.3-227.2)	2.2 (1.8 ï 2.5)	160.1 (128.2-182.0)

Table 1: Activities of antioxidant enzymes

* Patients with different diseases vs. CON; * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$; $^{+}$ CP vs. CHP, $^{+}$ $p < 0.05$, $^{++}$ $p < 0.01$

5. Disscusion

Although in all our studies increased oxidative stress was observed, the activities of measured antioxidant enzymes were not affected in all these diseases.

Catalase

Decreased activities of catalase were observed in patients with sepsis or septic shock, MetS and PC in comparison with CON, while in patients with DD, CP and AP no differences in CAT activity were detected.

The results of MetS studies previously published are inconsistent. In accordance with our results Koziróg et al. (2010) found decreased CAT activity in erythrocytes of MetS compared to CON. However no difference in erythrocyte CAT activity between MetS and CON were observed in two other studies (Broncel et al., 2010; Pizent et al. 2010). Cardona et al. (2008a, 2008b) found increased activities of serum CAT in MetS. Furthermore decreased activities of CAT were described in patients bearing only individual components of MetS ï obesity (Viroonudomphol, 2000), hypertension (Rodrigo, 2007) or insulin resistance (Shin, 2006).

Activities of CAT in erythrocytes were not altered in our set of DD women, in accordance with Bilici et al. (2001). However, Galecki et al. (2009) observed increased activity of CAT in erythrocytes in depressive patients compared to CON, Szuster-Ciesielska et al. (2008) found raised activities of CAT in serum of patients with major depression. In the study of patients with multiple sclerosis Miller et al. (2011) found elevated erythrocyte CAT activity in comparison with controls, regardless of the depression. Ozcan et al. (2004) described decreased CAT activities in erythrocytes of patients with affective disorders.

In the studies dealing with activities of CAT in SP the results are contradictory to our ones. Increased CAT activity was found in both erythrocytes and plasma (Warner et al., 1995) and also in serum (Leff et al., 1992 and Leff et al., 1993) of adult SP. Increased serum CAT activity was observed also in neonatal sepsis (Kapoor et al., 2006). Leff et al.

(1993) had reported that SP suffering from acute respiratory distress syndrome (ARDS) had higher activities of serum CAT than those SP without ARDS. However Metnitz et al. (1999) did not find any alterations in the erythrocyte CAT activity in patients with ARDS.
To the best of my knowledge, there is only one study dealing with the CAT activity in PC and in the contrast to our study, no changes in CAT activity were found (Fukui 2004).

No significant difference in CAT activities of CP patients observed in our study, were consistent with results of Fukui et al. (2004). In the contrast, other authors described increased (Szuster-Ciesielska, 2001a, Szuster-Ciesielska, 2001b) and also decreased (Quillot, 2005) CAT activities in patients with CP.

In accordance with our results no difference in erythrocyte CAT activity between AP and CON were found in study of Chmiel et al. (2002). It was shown, that serum CAT activity was higher in AP than in CON (Goth, 1989; Góth, 1982; Szuster-Czielska, 2001a; Fukui, 2004).

As mentioned above we found either decreased or unchanged activities of erythocyte CAT in observed disaeses. All these diseases were conected with increased oxidative stress as shown with elavated levels of CD-LDL and ox-LDL. Oxidative stress is caused by the imbalance between RONS production and degradation. It could be supposed, that in higher concentration of hydrogen peroxide, also the activity of CAT will be increased. It is known that CAT belongs to the most effective enzymes, the catalactic rate of catalase is among highest of known enzymatic rates (Kirkman et al., 2006). However it was previously shown that long-term exposure of CAT to H2O2 leads to the oxidation of the catalase bound NADPH to NADP+ and to a decrease in the initial activity of CAT about to 30 % of the initial activity (Kirkman et al., 1987).

Superoxide dismutase

Increased activities of CuZnSOD were observed in MetS, DD, PC and SP, while no difference in CuZnSOD activities were found between CP or AP and CON.

The raised CuZnSOD activities in the erythrocytes of patients with MetS found in our study may be compared with the results of Dimitrijevic-Sreckovic et al. (2007), who described slightly increased CuZnSOD activities in children with MetS in comparison with obese children without MetS. However the previously described results are not consistent, then in some studies decreased CuZnSOD activity in MetS patients were observed (Koziróg et al., 2010; Broncel et al., 2010) and in study of Pizent et al. (2010) no difference were found between MetS patients and CON. No difference between MetS and CON were also observed in EC-SOD activity (Cardona et al., 2008a).

We have found increased CuZnSOD activities in erythrocytes of DD compared with CON, similarly to Sarandol et al. (2007), Bilici et al. (2001), Gağecki et al. (2009) and Kotan et al. (2011). Inconsistent results were published for serum EC-SOD activities. Herken et al. (2007) and Selek et al. (2008) have found decreased, whereas Khanzode et al. (2003) and Szuster-Ciesielska et al. (2008) elevated EC-SOD activities in patients with major depression.

In line with our results of the study with patients with sepsis, Warner et al. (1995) found the increased activity of CuZnSOD at the onset of sepsis. On the other hand no differences in CuZnSOD activity between septic and healthy children (Cherian et al., 2007) as well as in patients with ARDS were detected (Metnitz et al., 1999). Also the activities of EC-SOD were found to be elevated in septic neonates compared to CON (Batra et al., 2000; Kapoor et al., 2006). Mühl et al. (2011) did not find any difference in EC-SOD activity between adult septic patients and CON.

No significant difference in the activities of CuZnSOD in CP patients and controls, found in our study, were consistent with the study of Quillot et.al. (2005). On the other hand, decreased CuZnSOD activity in CP patients was found in the study of Girish et al. (2011). Inconsistent results concerning serum SOD activities in hereditary and alcohol-related

pancreatitis have been published. Some reports have described increased (Mathew et al., 1996) or decreased (Szuster-Ciesielska et al., 2001a) SOD activity and in some studies no difference in SOD activities were found (Quillot et al., 2005, Quilliot et al., 2001).

In the study with AP patients we didnôt find any difference between CuZnSOD activity in AP and CON. Furthermore the levels of CuZnSOD were stable in the course of AP. The published results about CuZnSOD activity in AP are inconsistent. Some studies observed increased (Chmiel et al., 2002) and some decreased (Abu-Hilal et al., 2006; Park et al., 2003) CuZnSOD activity in patients with mild and/or severe AP. For EC-SOD, increased levels in AP were described (Góth et al., 1982; Góth, 1989; Szuster-Czielska et al., 2001a; Thareja et al., 2009).

Paraoxonase

The arylesterase activity of PON1 was measured in all observed pathophysiological states and was found to be decreased in all these situations (with the exception of DD) in comparison with CON. In patients with PC, CP and AP also the lactonase activity of PON1 was determined. In PC, CP and AP patients also PON1-L activity was found to be depressed compared with CON. As for paraoxonase activity of PON1, it was established only in the pilot study with septic patients. In septic patients lower activity of PON1-P then in CON was found. The PON1-P activity was not determined more because paraoxon which is used as substrate belongs to carcinogens.

The finding of decreased PON1-A in our subjects with MetS is in accordance with other studies (Hashemi et al., 2011; Kappelle et al., 2011; Martinelli et al., 2012). In patients with MetS were also found decreased PON1-P activities (Sentí et al., 2003; Garin et al., 2005; Rizos et al., 2005; Park et al., 2010; Hashemi et al., 2011; Akçay et al., 2011; Martinelli et al., 2012). However in studies of Tabur et al. (2010) and Lagos et al. (2009) equivalent levels of PON1-P and PON1-A in MetS patients and in CON were found. Also in study of Yilmaz et al. (2010) no

difference in PON1-P activities between MetS and CON were observed, however MetS patients with coronary artery disease (CAD) had significantly lower activity of PON1-P then MetS patients without CAD ($p < 0.008$). No difference in PON1-L activities between MetS and CON were observed (Martinelli et al., 2012).

No difference in PON1-A activities between DD and CON were found in our study in accordance with study of Sarandol et al. (2006). The published results are not consistent then also decreased levels of PON1-A activities in DD subjects were already described (Barim et al., 2009; Kotan et al., 2011). In all studies dealing with PON1-P activity equal levels of PON1-P were observed in DD subjects and CON (Sarandol et al., 2006; Barim et al., 2009; Kotan et al., 2011).

In both our studies, deeling with critically ill SP, we found lower PON1-A activity in in comparison with CON in accordance with the study of Draganov et al. (2010). Also PON1-P activities were found to be decreased in SP compared to CON in our pilot study. Similarly Kedage et al. (2010) observed decreased activity of serum PON1-P in septic patients.

We found decreased PON1-A and PON1-L activities in PC patients compared to CON. At the present time, the decreased PON1 activity in PC patients was described only in one study (Akçay et al., 2003a). However decreased PON1-A and/or PON1-P activities were observed in other malignancies such as in breast cancer (Samra et al., 2011), prostate cancer (Samra et al., 2011), lung cancer (Elkiran et al., 2007; Samra et al., 2011), laryngeal cancer (Karaman et al., 2010), endometrial cancer (Arioz et al., 2009), gastroesophageal cancer (Krzystek-Korpacka et al., 2008), gastric cancer (Akçay et al., 2003b) ovarian cancer (Camuzcuoglu et al., 2009), cervix carcinoma (Samra et al., 2011), lymphoma (Samra et al., 2011), high grade gliomas (Kafadar et al., 2006) or meningiomas (Kafadar et al., 2006). There is no study dealing with PON1-L activities in patients with any type of cancer.

The activity of PON1-A was established in experimental AP on animal model (36 male Wistar rats). The pancreatitis was induced by retrograde infusion into the biliopancreatic duct of 5% sodium taurocholate (severe AP) or of 1% sodium taurocholate (mild AP).

Control animals received an intraductal infusion of saline solution (0.9% NaCl). In this study they found no changes in PON1-A activity 3 hours after AP induction, by contrast after 18 hours after AP induction they observed significant decrease in PON1-A activity in severe AP compared to controls (Franco-Pons et al., 2008).

In all studies, where two different activities of PON1 were established, both PON1 activities correlated with each other. PON1-A activities correlated positively with PON1-P activities in septic patients and with PON1-L activities in patients with PC, CP and AP in our studies. Positive correlations between PON1-P and PON1-A were previously described in patients with MetS (Tabur et al., 2010; Hashemi et al., 2011) or with ovarian cancer (Camuzcuoglu et al., 2009).

Because PON1 is carried in plasma/serum bound to HDL through Apo-A1, the concentrations of HDL and Apo-A1 in serum and correlations between PON1 activities and concentrations of HDL and Apo-A1 were also established. In accordance with the results of PON1-A activity in patients with MetS, sepsis or septic shock, PC and AP decreased concentrations of both HDL and Apo-A1 were found. Positive correlation between PON1-A activity and HDL concentration was found in patients with DD, MetS and sepsis or septic shock. Positive correlation between PON1-A activity and Apo-A1 concentration was observed in patients with DD, MetS, sepsis or septic shock, CP and AP. It could be hypothesize, that changes in composition of HDL influence the activity and function of PON1.

Previously was shown that during the acute phase response, HDL structure is modified. It loses esterified cholesterol, apo-A1, and most of the HDL-associated enzymes including PON1 and that afterwards PON1 is replaced by SAA. These changes lead to the loss of HDL antioxidative properties (Van Leeuwen, 2003). The relationship between SAA concentrations and PON1 activities was also demonstrated in patients with MetS (Kappelle et al., 2011).

The levels of SAA were investigated in our studies with CP, PC and SP. Patients with PC and SP had higher concentrations of SAA than CON, while the SAA levels in CP were equal to those of CON. In these studied groups no correlations between PON1 activities and levels of

SAA were observed. The finding of increased SAA levels in PC patients in our study is consistent with results of other studies (Yokoi et al., 2005; Firpo et al., 2009). SAA was associated with tumour progression and its metastasizing (Malle et al., 2009). Some authors considered SAA as a tumour marker for PC, however, SAA did not reach appropriate specificity and sensitivity for PC diagnostics (Yokoi et al., 2005; Firpo et al., 2009). Elevated levels of SAA in sepsis were also demonstrated previously (Eras et al, 2011; Cetinkaya et al., 2009; Arnon et al., 2007).

Severeal mechanisms are supposed to decrease PON1 activity. It was shown, that increased oxidative stress connected with elevated levels of oxidized LDL cause inactivation of PON1. Oxidized LDL appears to inactivate PON1 through interactions between the enzymeôs free sulfhydryl group and oxidized lipids, which are formed during LDL oxidation (Aviram et al., 1999). Hydroperoxides of LA inhibit the PON1 activity through the reaction with sulfhydryl group of its cystein 284 (Tavori et al., 2011). Other reason for the decrease in PON1 activity could be the glycation of the enzyme, which takes place as was shown in diabetes mellitus (Hedrick et al., 2000). The acute phase response could also lead to the decreased activities of PON1 which are caused by the down-regulation of liver PON1 mRNA (Deakin & James, 2004). In the model of experimental acute pancreatitis was shown that the decrease of PON1 activities is connected with its inhibition by oxidized lipids and higher proteolytic degradation (Franco-Pons et al., 2008).

6. Conclusions

This doctoral thesis was focused on the behaviour of the antioxidant enzymes ï catalase, superoxide dismutase and paraoxonase in different pathophysiological states. Activities of these enzymes were investigated in patients with metabolic syndrome, depressive disorder, sepsis, pancreatic cancer, chronic, and acute pancreatitis.

In patients with metabolic syndrome activities of all three enzymes were altered in comparison with healthy controls. The erythrocyte activities of CuZnSOD were elevated and activities of CAT in erythrocytes and PON1-A in serum were decreased in the MetS patients.

In women with depressive disorders only erythrocyte activities of CuZnSOD were increased compared to controls. Activities of CAT and PON1-A were not altered in DD women.

Patients with sepsis had elevated levels of CuZnSOD activities and decreased activities of CAT and PON1-A and PON1-P compared to controls.

Patients with pancreatic carcinoma had elevated erythrocyte activities of CuZnSOD and decreased activities of CAT, PON1-A and PON1-L in comparison with controls. In patients with acute and chronic pancreatitis only activities of PON1 were depressed compared to controls. In activities of CAT and CuZnSOD no difference between AP or CP and CON were observed.

It was also shown that the different types of paraoxonase activities correlate with each other.

Our studies show that in pathophysiological states the activity of CuZnSOD is elevated and activities of catalase and paraoxonase are depressed when changed.

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