Journal of Microbiology, Biotechnology and Food Sciences

Petruška et al. 2012 : 2 (2) 510-516

REGULAR ARTICLE

EFFECT OF QUERCETIN AND T-2 TOXIN ON ANTIOXIDANT PARAMETERS OF PORCINE BLOOD *IN VITRO*

Peter Petruška[1], Anna Latacz[2], Adriana Kolesárová [1], Marcela Capcarová[1]*

Address: [1]Slovak University of Agriculture, Faculty of Biotechnology and Food Sciences, Department of Animal Physiology, Tr. A. Hlinku 2, 949 76 Nitra, Slovak republic

[2]University of Agriculture in Krakow, Faculty of Animal Sciences, Department of Animal Physiology and Endocrinology, Al. Mickiewicza 24/28, 54 137, Poland

*Corresponding author: petruska.peter85@gmail.com, phone number: 037/6414 4288

ABSTRACT

T-2 toxin, a trichothecene mycotoxin, is considered to be a one of the most toxic compounds that is produced by molds, particularly the *Fusarium* species. Mycotoxins can contaminate a large variety of feed mixtures, and could cause serious health problems to domestic livestock and humans when consumed. The aim of the present study was to investigate the effect of quercetin and T-2 toxin on some antioxidants parameters (superoxide dismutase - SOD, glutathione peroxidase - GPx) in porcine blood *in vitro*. Application of quercetin in different doses (1, 10, 100 $mg.ml^{-1}$) and T-2 toxin (1000 $ng.ml^{-1}$) has caused significant decrease of SOD activity. Differences among the groups in the case of GPx activity remained insignificant ($P<0.05$). In this study the additions of quercetin and T-2 toxin in different doses in porcine blood *in vitro* caused significantly ($P<0.05$) lower SOD activity in all experimental groups in comparison with the control group.

Keywords: T-2 toxin, quercetin, SOD, GPx, porcine blood

INTRODUCTION

Mycotoxins are natural and very stable toxins, with relatively low-molecular weight secondary metabolites of fungal origin, which can contaminate a large variety of feed mixtures **(Labuda *et al.*, 2009; Tančinová and Labuda, 2009)** grains and foodstuffs worldwide, a variety of foods and beverages, including both plant-based products and animal products **(Schollenberger *et al.*, 2007; Ranzenigo *et al.*, 2008)**.

Trichotecene mycotoxins are very large family of chemically related toxins produced by various species of *Fusarium, Myrotecium, Trichoderma, Cephalosporium,* etc. **(Wannemacher and Neufeld, 1991)**. T-2 toxin is some of the most important and toxic trichothecene mycotoxin occurring in various agriculture products **(Iwahashi *et al.*, 2008)**. Lipophilic nature of T-2 toxin suggests that they are easily absorbed through skin, gut, and pulmonary mucosa **(Bunner and Morris, 1988)**. Trichotecene causes multiorgan effect including emesis, and diarrhea, weight loss, nervous disorders, cardiovascular alterations, immunodepression, hemostatic derangements, skin toxicity, and bone marrow damage **(Wannemacher and Neufeld, 1991)**.

Quercetin, a member of the flavonoids family, is one of the most prominent dietary antioxidants. It is ubiquitously present in foods including vegetables, fruit, tea and wine as well as countless food supplements and is claimed to exert beneficial health effects. This includes protection against various diseases such as osteoporosis, certain forms of cancer, pulmonary and cardiovascular diseases but also against aging. Especially the ability of quercetin to scavenge highly reactive oxygen species (ROS) such as peroxynitrite and the hydroxyl radical is suggested to be involved in these possible beneficial health effects **(Boots *et al.*, 2008).**

The aim of the present study was to investigate the effect of quercetin and T-2 toxin on antioxidants parameters (superoxide dismutase - SOD, glutathione peroxidase – GPx), in whole blood of porcine *in vitro*.

MATERIAL AND METHODS

The blood was obtained from sexually mature porcine Large white (APRC, Nitra). Quercetin (Q) (Sigma-Aldrich, Co, USA) was added to the blood samples at the doses: 1, 10 and 100 $mg.ml^{-1}$ alone or in combination with T-2 toxin (T-2) (Romer Labs Division Holding

GmbH, Tulln, Austria) at doses 1000 ng.ml^{-1}(Table 1). The blood samples without addition of quercetin or T-2 toxin served as the control group (C).

Table 1 Application of quercetin and T-2 toxin in porcine blood *in vitro*

Group	Quercetin (mg.ml^{-1})	T-2 toxin (ng.ml^{-1})
C	0	0
E1	1	0
E2	10	0
E3	100	0
E4	1	1000
E5	10	1000
E6	100	1000

Legend: C – control group, E1–E6 – experimental groups with various doses of quercetin alone or in combination with T-2 toxin.

The blood was incubated for 5 hours at 37 ^{o}C. Enzyme activity of SOD and GPx was assayed by spectrophotometric analysis (Genesys 10, Thermo Fisher Scientific Inc., USA), using commercial assay kit (Randox, Bratislava, Slovakia).

SAS software and Sigma Plot 11.0 (Jandel, Corte Madera, USA) were used to conduct statistical analyses. One-way ANOVA was used to calculate basic statistic characteristics and to determine significant differences among experimental and control groups. Data presented are given as mean and standard deviation (SD). Differences were compared for statistical significance at the level $P<0.05$.

RESULTS AND DISCUSSION

It is know that in general mycotoxins cause oxidative stress **(Sehata *et al.*, 2005)** and this leads to peroxidation of membrane lipids and β-oxidation of fatty acids **(Jaeschke *et al.*, 2002).** It has been shown that the administration of T-2 toxin caused a significant increase in respiratory burst of macrophages, which lead to the release of ROS into the blood stream **(Cooray and Johnsson, 1990).**

In this study we found significant decrease ($P<0.05$) in SOD activity after administration of quercetin and quercetin in combination with T-2 toxin in all observed

experimental groups (E1–E6) in comparison with the control group (C). The lowest activity of SOD was observed in E5 and E6 groups in comparison with the control group (C) where quercetin in combination with T-2 toxin was applicated, and experimental groups where quercetin in various doses (E1-E4) was applicated, but without statistical significance (P>0.05). It is known, that natural substances can cause changes in antioxidant status. **Seven *et al.* (2010**) found that vitamin C and propolis decreased the SOD activity and showed a tendency to reduce CAT (catalase) and GSH (glutathione) levels. In different study **Vilà *et al.* (2002)** found that α-tocopherol was significantly lower for the T-2 toxin treated mice.

On the contrary, in our previous research **(Petruška *et al.*, 2012)** T-2 toxin in a combination with resveratrol caused significantly (P<0.05) higher activity of SOD in the experimental groups against the control group. This higher level of SOD activity could be affected by using different substance as antioxidant agent. In the another *in vitro* study **(Petruška and Capcarová, 2012)** T-2 toxin caused insignificant (P>0.05) increase of SOD in all experimental groups in comparison with the control group.

Kolesarová *et al.* (2012) found that reproductive toxicity of animals induced by a mycotoxin – deoxynivalenol (DON) can be inhibited by a protective natural substance – resveratrol.

In the another study **Medvedova *et al.* (2011)** found that DON has direct effect on secretion of growth factor IGF-I and steroid hormone progesterone in porcine granulosa cells.

After application of quercetin and T-2 toxin the activity of GPx was slightly lower in all experimental groups (E1–E6) in comparison with the control group (C), however without statistical significance (P>0.05). GPx was slightly higher in the first experimental group (E1) in comparison with the experimental groups (E2-E6), but without significant confirmation (P>0.05). In this paper the application of T-2 toxin and quercetin has not caused the significant differences in observed parameters in contrast with the previous study **(Petruška *et al.*, 2012)**. Natural antioxidant quercetin had probably supportive effect on antioxidant parameters and is able to maintain the antioxidant status of animal cells.

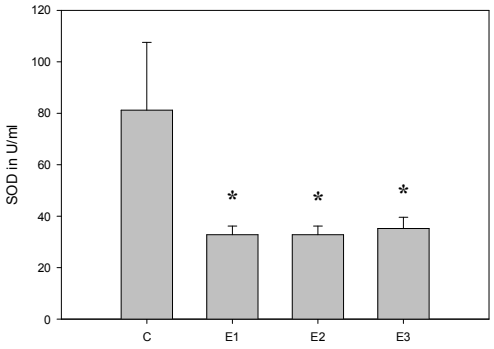

Figure 1 The activity of SOD of porcine blood after Quercetin exposure *in vitro*
C – control group
E1 - 1 mg.ml^{-1} Q
E2 – 10 mg.ml^{-1} Q
E3 – 100 mg.ml^{-1} Q
values are means ± SD
* means significant difference against control (P<0.05)

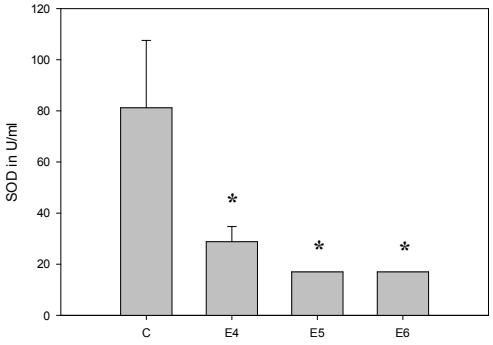

Figure 2 The activity of SOD of porcine blood after Quercetin and T-2 toxin exposure *in vitro*
C – control group
E4 - 1 mg.ml^{-1} Q + 1000 ng.ml^{-1} T-2
E5 – 10 mg.ml^{-1} Q + 1000 ng.ml^{-1}T-2
E6 – 100 mg.ml^{-1} Q + 1000 ng.ml^{-1} T-2
values are means ± SD
* means significant difference against control (P<0.05)

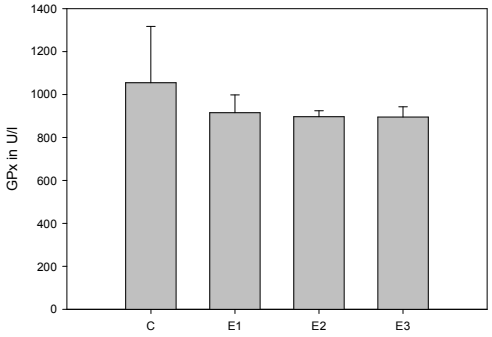

Figure 3 The activity of GPx of porcine blood after Quercetin exposure *in vitro*
C – control group
E1 - 1 mg.ml^{-1} Q
E2 – 10 mg.ml^{-1} Q
E3 – 100 mg.ml^{-1} Q
values are means ± SD

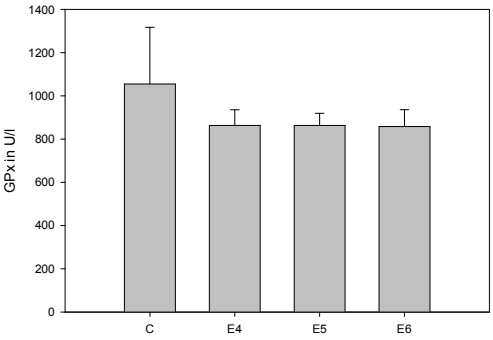

Figure 4 The activity of GPx of porcine blood after Quercetin and T-2 toxin exposure *in vitro*
C – control group
E4 - 1 mg.ml^{-1} Q + 1000 ng.ml^{-1} T-2
E5 – 10 mg.ml^{-1} Q + 1000 ng.ml^{-1}T-2
E6 – 100 mg.ml^{-1} Q + 1000 ng.ml^{-1} T-2
values are means ± SD

CONCLUSION

In this study the additions of quercetin and T-2 toxin in different doses in porcine blood *in vitro* caused some changes in antioxidant status. The activity of SOD was significantly (P<0.05) lower in all experimental groups in comparison with the control group. Also activity of GPx was slightly lower in the experimental groups but without significant differences (P>0.05). To our knowledge there are not a lot of similar studies on effect of quercetin and T-2 toxin in various doses given to the blood *in vivo* and its effect on antioxidant profile. Further investigation with different doses of T-2 toxin and combination with different doses of quercetin will be worthy of further investigation.

Acknowledgments: This work was financially supported by VEGA scientific grant 1/0790/11 and VEGA 1/0084/12.

REFERENCES

BOOTS, W. A. – HAENEN, R. M. M. G. – BAST, A. 2008. Health effects of quercetin: From antioxidant to nutraceutical. In *European Journal of Pharmacology*. vol. 585, 2008, no. 2-3, p. 325-337.

BUNNER, D. L. - MORRIS, E. R. 1998. Alteration of multiple cell membrane functions in L-6 myoblasts by T-2 toxin, an important mechanism of action. In *Toxicology and Applied Microbiology*. vol. 92, 1988, p. 113-121.

COORAY, R. - JOHNSSON, P. 1990. Modulation of resistence to mastitis pathogens by pretreatment of mice with T-2 toxin. In *Food Chemistry and Toxicology*. vol 28, 1990, p. 687-692.

IWAHASHI, Y. – KITAGAWA, E. – IWAHASHI, H. 2008. Analysis of mechanisms of T-2 toxin toxicity using yeast DNA Microarrays. In *International Journal of Molecular Sciences*. vol. 9, 2008, p. 2585-2600.

JAESCHKE, H. - GORES, G. J. - CEDERBAUM, A. I. - HINSON, J. A. - PESSAYRE, D. - LEMASTERS, J. J. 2002. Mechanisms of Hepatotoxicity. In *Toxicology Science*. vol. 65, 2002, p. 166-176.

KOLESAROVA, A. - CAPCAROVA, M. - MARUNIAKOVA, N. - LUKAC, N. - CIERESZKO, R. E. - SIROTKIN A. V. 2012. Resveratrol inhibits reproductive toxicity induced by deoxynivalenol. In *Journal of Enviromental Science and Health, Part A*. vol. 47, 2012 p. 1329-1334.

LABUDA, R. - PARICH, A. - VEKIRU, E. - TANČINOVÁ, D. 2009. Incident of fuminosis, moniliformin and Fusarium species in poultry feed mixtures from Slovakia. In *Annuals of Agricultural and Environmental Medicine*. vol. 12, 2009 p. 81-86.

MEDVEDOVA, M. - KOLESAROVA, A. - CAPCAROVA, M. - LABUDA, R. - SIROTKIN, V. A. - KOVACIK, J. - BULLA, J. 2011. The effect of deoxynivalenol on the secretion activity, proliferation and apoptosis of porcine ovarian granulosa cells *in vitro*. In *Journal of Environmental Science and Health, Part B*. vol. 46, 2011 p. 213-219.

PETRUŠKA, P. - CAPCAROVÁ, M. 2012. Effect of T-2 toxin on some parameters of antioxidant status of porcine blood *in vitro*. In *Animal physiology 2012*. Brno: Mendel University in Brno 2012, 2012. p. 157-161. ISBN 978-80-7375-616-1.

PETRUŠKA, P. - TUŠIMOVÁ, E. - CAPCAROVÁ, M. 2012. Effect of resveratrol and T-2 toxin on antioxidant parameters of porcine blood. *VI. Vedecká konferencia doktorandov s medzinárodnou účasťou*. Nitra : SPU v Nitre, 2012, p. 66-70.

RANZENIGO, G. – CALONI, F. - CREMONESI, F. - AAD, P. Y. - SPICER, L. J. 2008. Effect of *Fusarium* mycotoxins on steroid production by porcine granulose cells. In *Animal Reprodaction Science.* vol. 107, 2008, p. 115-130.

SCHOLLENBERGER, M. - MULLER, H. M. - RUFLE, M. - TERRY-JARA, H. - SUCHY, S. - PLANK, S. - DROCHNER, W. 2007. Natural occurrence of *Fusarium* toxins in soy food marketed in Germany. In *Internatiol Journal of Food Microbiology*. vol. 113, 2007, p. 142-146.

SEHATA, S. - KIYOSAWA, N. - ATSUMI, F. - ITO, K. - YAMOTO, T. - TERANISHI, M. - UETSUKA, K. - NAKAYAMA, H. - DOI, K. 2005. Microarray Analysis of T-2 Toxin - induced Liver, Placenta and Fetal Liver Lesions in Pregnant Rats. In *Experimental and Toxicologic Pathology*, vol. 57, 2005, p. 15-28.

TANČINOVÁ, D. - LABUDA, R. 2009. Fungi on wheat bran and their toxinogenity. In *Annuals of Agricultural and Environmental Medicine*, vol. 16, 2009, p. 325-331.

WANNEMACHER, R. W. JR. – BUNNER, D. L. – NEUFELD, H. A. 1991. Toxicity of trichothecenes and other related mycotoxins in laboratory animals. In *Smith JE, Henderson RS, eds. Mycotoxins and Animal Foods.* Boca Raton, Fla: CRC Press*;* 1991, p. 499–552.

WILSON, T. - KNIGHT, T. J. - BEITZ, D. C. - LEWIS, D. S. - ENGEN, R. L. 1996. Resveratrol promotes atherosclerosis in hypercholesterolemic rabbits. In *Life Sciences*. vol. 59, 1996, p. 15–21.