doi:10.4149/neo_2009_04_279

The Role of PPARs in MDR – a lesson from embryonic development
Minireview

A. KONIECZNA[1*], R. LICHNOVKA[1], B. ERDOSOVA[1], J. EHRMANN[1,2]

[1]Department of Histology and Embryology, Faculty of Medicine and Dentistry, Palacky University, Olomouc, e-mail: anusia@email.cz [2]Department of Pathology, Laboratory of Molecular Pathology, Palacky University, Olomouc, Czech Republic.

Received February 15, 2009

One of the most important features of embryonic cells is their resistence to xenobiotics, which provides a natural protection for embryos against these potentially harmfull molecules. In this way, embryo cells resemble cancer cells and thus understanding the basis of this phenomenon may contribute to overcoming the multi-drug-resistance (MDR) of some tumours. Peroxisome proliferator-activated receptors (PPARs) are steroid nuclear receptors that regulate diverse biological processes such as lipid and carbohydrate metabolism, development, differentiation, apoptosis, neoplastic transformation, inflammation and regeneration of tissues. Recently it has been found that they may also regulate the expression of some MDR proteins. In this article we summarise the main known relationships between some MDR pumps and three isoforms of PPAR receptors (PPAR-α, PPAR-β/δ, PPAR-γ). We hypothesize that regulation of MDR proteins by PPAR ligands in embryos could lead to the improvement of cancer treatment.

Key words: MDR, PPARs, embryogenesis, cancer

1. Introduction

Since the mid-19th century, our forebears in pathology noticed that tumors arise as embryo-like cells. This was based on the observed similarity between embryonal tissue and cancer tissue [1]. Cancer cells have reverted to an undifferentiated state and shared the features of embryo cells. Another hypothesis was that the real cause of tumor formation is to be sought in a defect or irregularity of the embryonic rudiment (Julius Cohnhein, 1889). The latter hypothesis was confirmed when stem cells emerged.

One important feature of embryonal cells is their resistence to xenobiotics, a feature which ensures the natural protection of the embryo against various external noxious substances. Therefore breaking down this resistance could contribute to overcoming the MDR which is now a major impediment in the successful treatment of cancer. In short, as embryo cells are similar to cancer cells, they may serve as a useful tool in the study of MDR.

2. Multi-drug-resistance (MDR)

Multi-drug-resistance (MDR) is now a serious obstacle in the treatment of patients with different types of tumors. MDR is the ability of tumor cells to resist the cytotoxic effects of a variety of structurally and functionally unrelated drugs. There are a large number of cellular factors contributing to drug resistance including the activation of de-toxifying enzymes, activation of DNA repair mechanisms, alterations in drug-induced apoptosis and increased drug efflux due to overexpression of membrane transporters [2]. In reality, drug resistance is not a single resistance mechanism but involves several simultaneously. Additionally, each tumor has its own unique resistance factor profile.

2.1. Multi-drug-resistance (MDR) pumps

MDR pumps, in other words, transmembrane proteins that belong to the ATP-binding cassette (ABC) transporter superfamily, play an important role in MDR. These use energy from ATP to transport various molecules across cell membranes. Their primordial role is to protect the organism from natural toxins by escorting them out of the cell. In cancer cells, MDR pumps cause drug resistence by the efflux of cell-killing chemotherapy molecules. These pumps are active in a broad spectrum of human cancers, including lymphoma, leukemia, breast, lung and ovarian cancers. The more MDR pumps cancer cells have the less effective chemotherapy is likely to be [3].

There are 48 human ABC transporters which are classified into seven distinct subfamilies of proteins called ABCA-G.

* Corresponding author

Three have been found in nearly all of MDR tumor cells in both human and rodent tissues. These include ABCB1/PGP/MDR1, ABCC1/MRP1 and ABCG2/MXR/BCRP [4]. The first is most often specified as P-glycoprotein (P-gp). It is a 170 kDa ATP-dependent efflux pump encoded by the *mdr1* gene [5, 6] located on chromosome 7q21.1 [7]. Mice have two homologues of ABCB1 (Abcb1a coding mdr1a and Abcb1b coding mdr1b). It is expressed in many normal tissues such as adrenal gland, kidney, liver, brain, intestine and hematopoietic stem cells where it has an excretory, protective or hormone handling role [8, 9]. Overexpression of this protein is sufficient to confer drug resistance [10]. It is possible to inhibit the function of P-gp by specific inhibitors but with the loss of its natural function. The second pump is known as multidrug resistance associated protein 1 (MRP1). It is located on chromosome 16p13.1. Yet, unlike P-gp, MRP1 operates with drugs or biologically active endogenous substrates conjugated with glucuronide, sulfate or glutathione [11]. Additionally, four homologues of MRP1 have been identified: MRP2/cMOAT (encoded by ABCC2), MRP3 (encoded by ABCC3), MRP4/MOATB (encoded by ABCC4) and MRP5/MOATC (encoded by ABCC5). Their physiological role is still unknown, however it is possible that they play role in cellular detoxification process and are involved in mediating drug resistence, too [12]. The third pump is located on chromosome 4q22 and has been identified in multidrug-resistant colon cancer cell line and breast cancer cell line that does not overexpress ABCB1 or ABCC1 [13]. Recently Krishnamurthy et al. suggested that expression of this pump is induced by hypoxia and this regulation involves the hypoxia-inducible transcription factor complex HIF-1 [14].

2.2 Expression patterns of MDR pumps in embryonal/fetal tissues

The main MDR pumps described above have also been found in embryonal/fetal tissues. A high level of mouse mdr1b was found in the endometrium and placental trophoblast of the pregnant uterus. It is assumed that its physiological role is to protect the fetus against xenobiotics during pregnancy [15]. P-gp is present in oocytes and early cleavage embryos where it mediates a self-protecting function during the time of germ cell maturation and early pre-implantatory development [16]. Kalken et al. studied the expression of P-gp in fetal tissues obtained during different developmental stages using immunohistochemistry. They compared the distribution of this protein in fetal and adult tissues. Differences were found in adrenal, intestine, respiratory epithelium, and brain capillaries [17]. Mdr1 mRNA begins to be expressed in the thymus of chicken embryo from day 12 until hatching but in the bursa from day 14 to day 17 of embryonic life [18]. Matsuoka et al. studied expression and localization of P-gp in the rat brain during development. It is localized in brain capillaries and was first detected on postnatal day 7 and then gradually increased to reach a plateau in the adult brain [19]. While the level of rat mdr1a and mdr1b transcript increases, the level of Mrp1 transcript is the same in all tissues during rat ontological development [20]. The high levels of ABCG2 in a subpopulation of hematopoetic stem cells [21] and the trophoblast cells of the placenta [22] suggest that it can transport compounds into fetal blood and remove toxic drugs and metabolites.

3. Multi-drug-resistance (MDR) and peroxisome proliferator-activated receptors (PPARs)

Recently a few studies have reported that the expression level of some MDR proteins can be regulated by activation or deactivation of PPAR receptors (see below).

3.1. Peroxisome proliferator-activated receptors (PPARs)

PPARs are steroid nuclear receptors which belong to the same superfamily as thyroid, retinoid and vitamin D receptors [23]. They regulate diverse biological processes such as lipid and carbohydrate metabolism, development, differentiation, apoptosis, neoplastic transformation, inflammation and the regeneration of tissues. Their ligands are thus becoming a good tool in the treatment of some serious human conditions such as obesity, type two diabetes, arteriosclerosis, infertility and cancer.

PPARs are transcription factors that regulate the expression of specific genes in a ligand-dependent manner. They were first described at the beginning of 1990's as the receptor activated by rodent hepatocarcinogens that cause peroxisome proliferation [24]. Three new members of these receptors (PPAR-α, PPAR-β/δ and PPAR-γ) were later found in the *Xenopus* frog and these had hypolipidemic effects during stimulation of peroxisomal degradation of fatty acids [25]. They require heterodimerization with the retinoid X receptor to start transcriptional activity [26]. They consist of 6 functional domains designated A to F. The N-terminal A/B region has a variable length and an autonomous activation function (AF-1). The most conserved C-domain is DNA-binding domain (DBD) that consists of two zinc-finger-like motifs characteristic for nuclear receptors. The D domain is a variable hinge. The multifunctional E domain encompasses the ligand-binding domain (LBD), a second activation function (AF-2) critical for transcriptional activation, a dimerization domain and region involved in nuclear localization. The F domain is present only in some nuclear receptors (absent in PPARs) [23]. In the presence of an agonist, DBD is bound to promoter sequences called the peroxisome proliferator response elements (PPRE) and this triggers expression of specific genes. PPREs are composed of two direct repetitions (DR-1) of the consensus sequence AGGTCA with a single nucleotide spacing between the two repeats [27]. Natural ligands of PPARs include fatty acids and eicosanoids. Synthetic ligands include lipid lowering drugs (like fibrates) and insulin sensitisers (like thiazolidinediones). PPARs do not contact the basal transcription machinery directly but require interaction with co-regulator complexes such as coactivator for stimulation or a co-repressor for inhibition of target gene expression [28].

3.2. PPAR isoforms and their localization

The PPAR subfamily consists of the three isoforms: PPAR-α, PPAR-β/δ and PPAR-γ. Each is coded by different genes located on different chromosomes. The receptors differ in their physiological roles, rate of expression in different tissues and ligand specificity. All three isotypes are widely expressed in somatic cells, in the developing fetus and in the germ cells of the ovary and testis. In rat PPAR-β/δ is expressed in the early phase of prenatal development with a marked peak at days 13,5-15,5, whereas PPAR-α and PPAR-γ appear later in the tissues where they will continue to be expressed in adulthood [30], with the exception of a transient expression in the CNS and the epidermis. PPAR-α was detected in tissues with a high catabolic rate (increased β-oxidation) of fatty acids and high peroxisome-dependent activities (liver, heart, kidney, intestine and brown adipose tissue) [31]. PPAR-β/δ has the broadest expression pattern. The levels of expression of this isotype depend on the extent of cell proliferation and differentiation. It has been found in skin, gut, placenta, skeletal muscle, adipose tissue and brain [32]. PPAR-γ has been the most studied. It is the most divergent isotype expressed as the two isoforms –γ1 and –γ2, that differ at their N terminus. Each isoform is expressed by a different promoter. PPARγ2 is found in high levels in different adipose tissues [33], whereas PPARγ1 has been detected in gut, brain, vascular cells and specific types of immune and inflammatory cells [34]. PPAR-γ has also been found in the granulosa cells that surround and support the maturing oocyte. This isotype is a negative regulator of follicular growth and differentiation. Its activation suppresses follicle development [35]. PPAR-β/δ and PPARγ have a crucial role in the placenta formation. Homozygote disruption of these two isotypes results in the death of the fetus by day 10 of development [36, 37].

4. The concept of the regulation of MDR expression by PPAR ligands

Generally the concept of the regulation of MDR expression by PPAR ligands has been postulated by several autors and here we summarize the main known relationships between these groups of proteins.

4.1. PPAR-α agonists and Bcrp/Abcg2, MRP1, Mdr1a

Hirai et al. compared the expression level of some ABC transporters after treating mice with a normal diet and diet containing PPAR-α agonists. The expression of Bcrp/Abcg2 was up-regulated, MRP1 down-regulated while the expression of Mdr1a remained unchanged [38].

4.2. PPAR-α agonist and MRP3, MRP4

Moffit et al. noticed increased levels of mRNAs and proteins of liver Bcrp, Mrp3 and Mrp4 in mice after clofibrate (PPAR-α agonist) treatment [39]. Also Maher et al. found that perfluorooctanoic acid and perfluorodecanoic acid (PFDA) increase expression of hepatic Mrp3 and Mrp4 mRNA. The presence

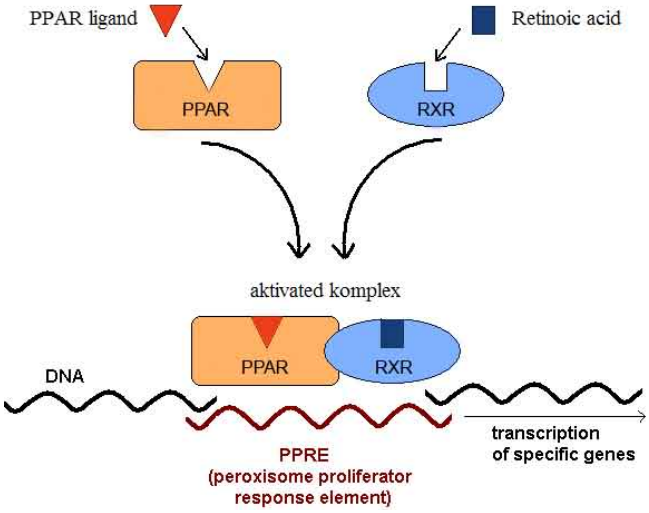

Figure 1. Mechanism of PPAR action – modified according to Štulc et al. [29].

of several direct repeat-1 elements (DR-1) of PPRE in the Mrp3 and Mrp4 5' regulatory regions suggest that PPAR-α may regulate these two transporters directly. MRP3 and MRP4 transporters can be also regulated by another transcription factor called NF-E2-related factor-2 (Nrf2). It is activated by PFDA directly or due to PPAR-α-dependent activation of drug-metabolizing enzymes [40].

4.3. PPAR-γ agonists and ABCG2

Szatmari et al. demonstrated that PPAR-γ directly and transcriptionally induces the ABCG2 expression in human monocyte-derived dendritic cells and myeloid leukemia cell line MM6. They identified and characterized a 150-base pair long conserved enhancer region, containing three functional PPAR response elements (PPRE), upstream of the human ABCG2 gene. All three PPRE contain direct repeat (DR-1) motifs and are able to at the end of the sentence bind PPAR-γ/RXR heterodimers [41].

4.4. PPAR-γ agonists and stroma

Hafner et al. supposed that PPAR-γ agonists influence stroma functions in cancer, too. They affect in particular angiogenesis but they also affect the immune response and they have direct anti-tumor effects [42].

4.5. TNF-α and MDR1, PPAR-α

Wang et al. noticed that tumor necrosis factor-α (TNF-α) could induce down-regulation of MDR1 and up-regulation of PPAR-α and in this way enhance cytotoxicity by apoptosis in HepG2/ADM cells [43].

4.6. PPARs and Hedgehog (Hh) signalling pathway

However, not all above mentioned relationships explain signalling crosstalk between MDR/PPARs pathways. Based on a few

studies we hypothesize that Hedgehog (Hh) signalling pathway may connect these two mentioned upwards. Sims-Mourtada noticed that Hh pathway activation induces expression of two ABC transporter proteins: ABCB1/PGP/MDR1 and ABCG2/MXR/BCRP [44]. Varnat et al. noticed that PPAR-β/δ regulate Paneth cell differentiation and homeostasis by down-regulating the expression of Indian hedgehog (Ihh) [45]. Additionally, Fontaine et al. found that Hedgehog signalling pathway control adipocyte maturation by targeting PPAR-γ2 expression [46]. Also Kim et al. noticed that 20(S)-hydroxycholesterol inhibit adipogenic differentiation through a hedgehog-dependent mechanism and this is associated with inhibition of PPARγ expression [47].

5. Conclusion

Based on these studies, we suggest that a detailed description of MDR regulation in embryos with particular focus on MDR/PPAR pathways could provide us with important information enabling us to target MDR in cancers using synthetic PPAR ligands.

Acknowledgements. We thank our technicians: Mrs. Eva Kašpárková and Mrs. Eva Nezhybová for their excellent assistances. This study was supported by MSM 6198959216.

References:

[1] Recamier JCA. Recherches dur le Traitement du Cancer, etc. Paris. 1829. Editorial Archiv fuer pathologische Anatomie und Physiologie und fuer klinische Medizin 8 (1855) 23.

[2] Gottesman MM. Mechanisms of cancer drug resistence. Annu Rev Med 2002; 53: 615–27. doi:10.1146/annurev.med.53.082901.103929

[3] Davis Alison. Tiny pumps protect many animals from toxins – sometimes a little too well. Science Notes 1997.

[4] Dean M. The Human ATP-Binding Cassette (ABC) Transporter Superfamily. J Lipid Res 2001; 42: 1007–17.

[5] Ueda K, Clark DP, Chen C et al. Expression of full-length cDNA for the human MDR1 gene confers resistence to kolchicine, doxorubicin, and vinblastine. Proc Nat Acad Sci 1987; 84: 3004–8. doi:10.1073/pnas.84.9.3004

[6] Sakaeda T, Nakanuta T, Okumura K. MDR1 genotype-ralated pharmacokinetics and pharmacodynamics. Biol Pharm Bull 2002; 25: 1391–1400. doi:10.1248/bpb.25.1391

7] Callen DF, Baker E, Simmers RN et al, Localization of the human multiple drug resistance gene, MDR1, to 7q21.1. Hum Genet 1987; 77: 142–4. doi:10.1007/BF00272381

[8] Thiebaut F, Tsuruo T, Hamada H et al. Cellular localization of the multidrug-resistance gene product P-glycoprotein in normal human tissues. 1987; 84: 7735–8.

[9] Fojo AT, Ueda K, Slamon DJ et al. Expression of a multidrug-resistance gene in human tumors and tissues. Proc. Nat. Acad. Sci 1987; 84: 265–9. doi:10.1073/pnas.84.1.265

[10] Gros P, Neriah YB, Croop JM et al. Isolation and expression of a complementary DNA that confers multidrug resistance. Nature 1986; 323(6090): 728–31. doi:10.1038/323728a0

[11] Ishikawa T, Kuo MT, Furuta K et al. The human multidrug resistance-associated protein (MRP) gene family: from biological function to drug molecular design. Clin Chem Lab Med 2000; 38: 893–7. doi:10.1515/CCLM.2000.130

[12] Kool M, Haas M, Scheffer GL et al. Analysis of expression of cMOAT (MRP2), MRP3, MRP4 and MRP5, homologues of the multidrug resistence-associated protein gene (MRP1), in human cancer cell lines. Cancer Res 1997; 3537–47.

[13] Miyake K, Mickley L, Litman T et al. Molecular cloning of cDNAs which are highly overexpressed in mitoxantrone-resistant cell: demonstration of homology to ABC transport genes. Cancer Res 1999; 59: 8–13.

[14] Krishnamurthy P, Ross DD, Nakanishi T et al. The stem cell marker Bcrp/ABCG2 enhances hypoxic cell survival through interactions with heme. J Biol Chem 2004; 279: 24218–25. doi:10.1074/jbc.M313599200

[15] Gottesman MM. How Cancer Cells Evade Chemotherapy: Sixteenth Richard and Hinda Rosenthal Foundation Award Lecture. Cancer Res 1993; 15; 53: 747–54.

[16] Elbling L, Berger W., Rehberger A,et al. P-glycoprotein regulates chemosensitivity in early developmental stages of the mouse. FASEB J 1993; : 1499–506.

[17] Kalken CK, Giaccone G, Valk P et al. Multidrug resistence gene (P-glycoprotein) expression in the human fetus. American Journal of Patology 1992; 141: 1063–72.

[18] Petrini M, Galimberti S, Sabbatini A et al. Expression of multidrug resistance (mdr) gene(s) in primary lymphoid organs of chicken immune system during embryonic development. Experienta 1995; 15; 51: 137–40. doi:10.1007/BF01929357

[19] Matsuoka Y, Okazaki M, Kitamura Y et al. Developmental expression of P-glycoprotein (multidrug resistance gene product) in the rat brain. J Neurobiol 1999; 5; 39 : 383–92. doi:10.1002/(SICI)1097-4695(19990605)39:3<383::AID-NEU5>3.0.CO;2-4

[20] Rosati A, Maniori S, Decorti G et al. Physiological regulation of P-glycoprotein, MRP1, MRP2 and cytochrome P450 3A2 during rat ontogeny. Dev Growth Differ 2003; 45: 377–87. doi:10.1046/j.1440-169X.2003.00699.x

[21] Zhou S, Schuetz JD, Bunting KD et al. The ABC transporter Bcrp1/ABCG2 is expressed in a wide variety of stem cells and is a molecular determinant of the side-population phenotype. Nat Med 2001; 7: 1028–1034. doi:10.1038/nm0901-1028

[22] Rocchi E, Khodjakov A, Volk EL et al. The product of the ABC half-transporter gene ABCG2 (BCRP/MXR/ABCP) is expressed in the plasma membrane. Biochem Biophys Res Commun 2000; 271: 42–46. doi:10.1006/bbrc.2000.2590

[23] Robyr D, Wolffe AP, Wahli W. Nuclear hormone receptor coregulators in action: diversity for shared tasks. Molecular Endocrinology 2000; 14 : 329–347. doi:10.1210/me.14.3.329

[24] Issemann I, Green S. Activation of a member of the steroid hormone receptor superfamily by peroxisome proliferators. Nature 1990; 18: 347(6294): 645–50.

[25] Dreyer C, Krey G, Keller et al. Kontrol of the peroxisomal β-oxidation pathway by a novel family of nuclear hormone receptors. Cell 1992; 6; 68: 879–87. doi:10.1016/0092-8674(92)90031-7

[26] Kliewier SA, Umesono K, Noonan DJ et al. Convergence of 9- cis retinoic acid and peroxisome proliferator signalling pathways

through heterodimer formation of their receptors. Nature 1992; 358(6389): 771–4. doi:10.1038/358771a0

[27] Schachtrup Ch, Emmler T, Bleck B et al. Functional analysis of peroxisome-proliferator-responsive element motifs in genes of fatty acid-binding proteins. Biochem. J. 2004; 382, 239–45. doi:10.1042/BJ20031340 PMid:15130092 PMCid:1133936

[28] Yu Sontago, Reddy JK. Transcription coactivators for peroxisome proliferator-activated receptors. Biochimica et Biophysica Acta 2007; 1771: 936–51.

[29] Štulc T, Češka R. Dual blockade of PPAR receptors. Farmakoterapie 2006; 1 (in Czech).

[30] Braissant O, Foufelle F, Scotto C et al. Differential expression of peroxisome proliferator-activated receptors (PPARs): tissue distribution of PPAR-alpha, -beta and –gamma in the adult rat. Endocrinology 1996; 137: 354–366. doi:10.1210/en.137.1.354

[31] Mandart S, Muller M, Kersten S. Peroxisome proliferators-activated receptor alpha target genes. Cell Mol Life Sci. 2004; 61: 393–416. doi:10.1007/s00018-003-3216-3

[32] Willson, TM., Brown, PJ., Sternbach, DD. The PPARs: From orphan receptors to drug discovery. J Med Chem 2000; 43(4): 528–550. doi:10.1021/jm990554g

[33] Chabla A, Schwarz EJ, Dimaculangan DD et al. Peroxisome proliferator-activated receptor (PPAR) gamma: adipose predominant expression and induction early in adipocyte differentiation. Endokrinology 1994; 135: 798–800. doi:10.1210/en.135.2.798

[34] Zhu Y, Qi C, Kornberg JR et al. Structural organization of mouse peroxisome proliferator-activated receptor gamma (mPPAR-gamma) gene: alternative promoter use and different splicing yield two mPPAR gamma isoforms. Proc Natl Acad Sci USA 1995; 92: 7921–7925. doi:10.1073/pnas.92.17.7921

[35] Lovekamp-Swan T, Chaffin CL. The peroxisome proliferation-activated receptor gamma ligand troglitazone induces apoptosis and p53 in rat granulose cells. Molecular and Cellular Endocrinology 2005; 15;233: 15–24. doi:10.1016/j.mce.2005.01.011

[36] Nadra K, Anghel SI, Joye E. Differentiation of trophoblast giant cells and their metabolic functions are dependent on peroxisome proliferator-activated receptor beta/delta. Molecular and Cellular Biology 2006; 26: 3266–81 doi:10.1128/MCB.26.8.3266-3281.2006

[37] Barak Y, Nelson MC, Ong ES. PPARγ is required for placental, cardiac and adipose tissue development. Molecular Cell. 1999; 4: 585–95. doi:10.1016/S1097-2765(00)80209-9

[38] Hirai T, Fukui Y, Motojima K. PPARalpha agonists positively and negatively regulace the expression of several nutriet/drug transportere in mouse small intestine. Biol Pharm Bull 2007; 30: 2185–90. doi:10.1248/bpb.30.2185

[39] Moffit JS, Aleksunes LM, Maher JM et al. Induction of hepatic transporers multidrug resistence-associated proteins (Mrp) 3 and 4 by clofibrate is regulated by peroxisome proliferator--activated receptor alpha. J Pharmacol Exp Ther 2006; 317: 537–45. doi:10.1124/jpet.105.093765

[40] Maher JM, Aleksunes LM, Dieter MZ et al. Nrf2- and PPAR alpha--mediated regulation of hepatic Mrp transporters after exposure to perfluorooctanoic acid and perfluorodecanoic acid. Toxicol Sci 2008; 106: 319–28. doi:10.1093/toxsci/kfn177

[41] Szatmari I, Vámosi G, Brazda P et al. Peroxisome proliferator--activated receptor gamma-regulated ABCG2 expression confers cytoprotection to human dendritic cells. 2006; J Biol Chem 281: 23812–23. doi:10.1074/jbc.M604890200

[42] Hafner C, Reichle A, Vogt T. New indications for established drugs: combined tumor-stroma-targeted cancer therapy with PPARgamma agonists, COX-2 inhibitors, mTOR antagonists and metronomic chemotherapy. Curr Cancer Drug Targets 2005; 5: 393–419. doi:10.2174/1568009054863591

[43] Wang Q, Chen XP, Hai S et al. TNF-alpha induced reversal of multidrug resistence in human hepatocellular carcinoma cells. Zhonghua Wai Ke Za Zhi 2007; 1; 45: 602–4.

[44] Mourtada JS, Izzo JG, Alani J et al. Sonic hedgehog promotes multiple drug resistence by regolation of drug transport. Oncogene 2007; 16;26: 5674–9. doi:10.1038/sj.onc.1210356

[45] Varnat F, Ten Heggeler BB, Grisel P et al. PPARbeta/delta regulates panech cell differentiation via controlling the hedgehog signaling pathway. Gastroenterology 2006; 131: 538–53. doi:10.1053/j.gastro.2006.05.004

[46] Fontaine C, Cousin W, Plaisant M et al. Hedgehog signaling alters adipocyte maturation of human mesenchymal stem cells. Stem Cells. 2008; 26: 1037–46. doi:10.1634/stemcells.2007-0974

[47] Kim WK, Meliton V, Amantea CM et al. 20(S)-hydroxycholesterol inhibits PPARgamma expression and adipogenic differentiation of bone marrow stroma cells through a hedgehog-dependent mechanism. J Bone Miner Res 2007; 22: 1711–9. doi:10.1359/jbmr.070710