Involvement of the L-cysteine/CSE/H_2S pathway in human melanoma progression

C. Armogida[1], E. Panza[1], P. De Cicco[1], G. Ercolano[1], G. Botti[2], A. Papapetropoulos[3], G. Cirino[1], A. Ianaro[1]

[1]Dept. of Pharmacy, University of Naples Federico II, Naples, Italy
[2]Unit of Pathology, Istituto Nazionale per lo Studio e la cura dei tumori 'Fondazione G. Pascale' IRCCS, Naples, Italy
[3]Dept. of Pharmacy, University of Athens, Athens, Greece

Metastatic melanoma is an aggressive disease with a median survival of less than 1 year. Although dramatic improvements have been made in the last two years with the approval of several new drugs for the treatment of metastatic melanoma, most responses of these drugs are partial and relapse of the disease appears in 5-7 months. In the last few years, numerous physiological and pathophysiological roles have been proposed for the gasotransmitter hydrogen sulfide (H_2S), along with a plethora of cellular and molecular targets. Endogenously, H_2S is produced as a metabolite of homocysteine (Hcy) by cystathionine β-synthase (CBS), cystathionine γ-lyase (CSE), and 3-mercaptopyruvate sulfurtransferase (3MST) (Sen et al., 2012). A number of studies have investigated the role of H2S in triggering cell death and evidence has been presented that this gas can exert both pro- and anti-apoptotic activity in cultured cells (Hu et al., 2007) (Taniguchi et al., 2011). The aim of our study was to evaluate the role of the metabolic H_2S pathway in human melanoma. Using immunohistochemistry we demonstrated the presence of the L-cysteine/CSE/H_2S pathway in human melanoma specimens and provided evidence that CSE expression was highest in primary tumors while decreased in the metastatic lesions and it was almost silent in non lymph node metastases. Conversely, CBS did not appear to play an important role in melanoma. The role of CSE and the downstream signal transduction were investigated by using several human melanoma cell lines. The primary role played by CSE, already revealed by the human study, was confirmed by the finding that the over-expression of CSE induced spontaneous apoptosis of human melanoma cells (30%; *P<0.001 vs* mock transfected cells). The same effect was achieved by using different H_2S donors and, among them, the most active resulted to be diallyl trisulfide (DATS) (IC_{50}= 89 μM). The main pro-apoptotic mechanisms involved were suppression of nuclear factor-κB activity and inhibition of AKT and ERK1/2 pathways. A proof of concept was obtained *in vivo* by using a murine melanoma model. In fact, either L-cysteine, the CSE substrate, or DATS inhibited tumor growth by 51% and by 67% respectively *(P*<0.001, n=8). In conclusion, this work establishes that the L-cysteine/CSE/H_2S pathway is involved in human melanoma and provide the fundamentals to exploit a possible therapeutic/diagnostic use in this aggressive disease.

Hu et al. (2007). *J Neurochem.*100:1121–1128.
Sen et al. (2012). *Am J Physiol Cell Physiol*. 303(1):C41-51.
Taniguchi et al. (2011).*Br J Pharmacol*. 162:1171–1178.