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PERSPECTIVES

Necrotic amplification loop in acute pancreatitis: pancreatic stellate cells and nitric oxide are important players in the development of the disease

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Acute pancreatitis (AP) is a serious, life-threatening disease with high mortality. Unfortunately, there is still no specific therapy, and therefore the task of finding one remains with basic researchers. The exocrine pancreas contains many cell types, including pancreatic acinar cells (PACs), pancreatic ductal cells (PDCs), inflammatory cells (ICs) and pancreatic stellate cells (PSCs). PACs, PDCs and ICs are usually blamed for acute inflammation, while PACs, PDCs and PSCs are thought to be responsible for chronic inflammation. The generally accepted theory of AP is that (1) toxic factors (such as alcohol, bile or fatty acids) induce intracellular calcium signalling, mitochondrial damage, depletion of both glycolytic and oxidative ATP synthesis, and ER stress in PACs and PDCs; (2) this is followed by resultant intra-acinar and luminar trypsinogen activation and fluid and bicarbonate secretory deficit; (3) the continuous decrease of pH enhances the autoactivation of trypsinogen, leading in turn to cell death (Pallagi *et al.* 2011); and (4) this latter mechanism will then attract the inflammatory cells to the pancreas and elevate the cytokine level, spreading the local necrosis and causing a serious systemic necroinflammatory disease (Hegyi & Petersen, 2013).

In this issue of *The Journal of Physiology*, Gryshchenko *et al.* (2018) describe new mechanisms which add a very important piece to the puzzle of the AP pathomechanism. The authors very elegantly record Ca^{2+} signalling in different cell types in the exocrine pancreatic lobules. They clearly show that it is not only PACs and PDCs that can respond to various stimuli, but PSCs as well. Notably, PSCs could have been triggered to evoke intracellular Ca^{2+} by physiological (ATP, bradykinin, vasoactive intestinal peptide and bombesin) and pathophysiological (ethanol and fatty acids) stimuli, but not by membrane depolarization or trypsin. This information would not be surprising alone, but this pattern totally changes during acute pancreatitis. The authors show that the responsiveness of PSCs to physiological stimuli (bradykinin) decreases in the ethanol–fatty acids pancreatitis model, while PSCs become very sensitive to trypsin. Notably, administration of trypsin induced nitric oxide (NO) formation and a Ca^{2+} signal in PSCs (Jakubowska *et al.* 2016). NO then diffuses into adjacent PACs and contributes to further damage to PACs. It must be noted that PAC necrosis elevates

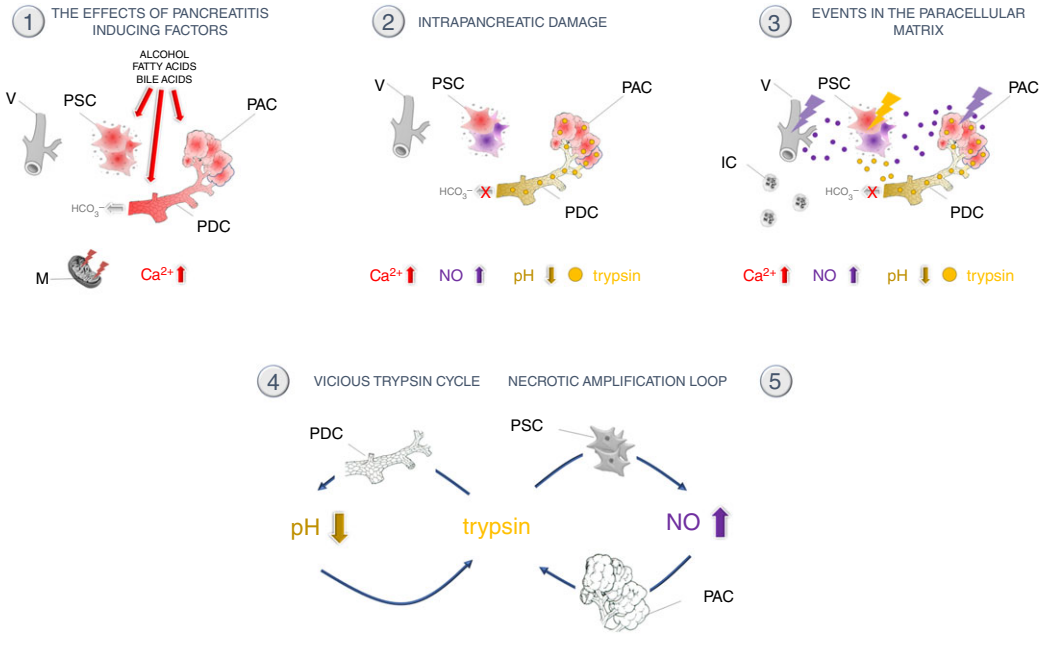

Figure 1. Pathomechanism of acute pancreatitis
IC, pancreatic inflammatory cell; M, mitochondrion; NO, nitric oxide; PAC, pancreatic acinar cell; PDC, pancreatic ductal cell; PSC, pancreatic stellate cell; V, blood vessel.

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the bradykinin level, which can stimulate NO formation and Ca^{2+} signals in PSCs. This necrotic amplification loop between PACs and PSCs has serious consequences in AP, since the cells continuously trigger and damage each other without intervention (Jakubowska *et al.* 2016; Gryshchenko *et al.* 2018).

The discovery of the necrotic amplification loop also helps to answer the question of the source of the elevated nitrite/nitrate (NO_x) level in AP. NO_x levels significantly increase in the blood and in the lungs in cerulein-, ethanol-, pancreatic duct obstruction- and taurocholate-induced experimental AP models. Moreover, supramaximal doses of cerulein and injection of ethyl alcohol into the pancreatic duct significantly elevate the pancreatic contents of NO_x (Hegyi & Rakonczay, 2011). Although almost all authors to date have confirmed that the increased serum NO_x levels most probably originated from non-acinar cell types, it is Petersen's workgroup who have shown that PSCs are at least in part responsible for the elevated NO_x level (Jakubowska *et al.* 2016; Gryshchenko *et al.* 2018).

Importantly, the elevated NO_x level not only damages PACs, but also decreases the velocity of the pancreatic microcirculation and elevates the number of adherent leukocytes in the pancreas (Hegyi & Rakonczay, 2011). The fact that inhibition of the inducible NO synthase improves outcomes in experimental AP models and that pharmacological inhibition of NO synthase provides remarkable protection against necrosis confirms the possibility of drug development against the necrotic amplification loop (Hegyi & Rakonczay, 2011; Jakubowska *et al.* 2016). Since many other vicious cycles and loops can be found inside the pancreas during AP, a complex understanding of how the disease develops is crucial.

Therefore, Gryshchenko *et al.*'s article changes our understanding of the pathomechanism of AP (Fig. 1) as follows:

(1) Toxic factors (i.e. ethanol, fatty acids and bile) induce a sustained Ca^{2+} signal in PACs, PSCs and PDCs.
(2) Fluid and bicarbonate secretion is blocked in PDCs, pH decreases in the pancreas and pancreatic lumen, trypsinogen is activated in PACs, and NO is synthesized in PSCs.
(3) NO damages PACs, elevating the amount of trypsin in the paracellular matrix; decreases the velocity of the pancreatic microcirculation; and elevates the level of inflammatory cells.
(4) Trypsin further inhibits PDCs by inhibiting cystic fibrosis transmembrane conductance regulator (CFTR) (Pallagi *et al.* 2011)
(5) Trypsin stimulates NO production in PSCs (Gryshchenko *et al.* 2018).

The resultant necrosis will then attract the inflammatory cells to the pancreas and elevate the cytokine level, spreading the local necrosis and thus causing a serious systemic necro-inflammatory disease.

Both the vicious trypsin cycle (Pallagi *et al.* 2011) and the necrotic amplification loop (Gryshchenko *et al.* 2018) must be blocked to develop a specific therapy for AP. Because of the multifactorial pathomechanism, several combinations of CFTR correctors/activators, bradykinin receptor antagonists, protease-activated receptor antagonists, nitric oxide synthase inhibitors, store-operated Ca^{2+} entry blockers and mitochondrial permeability transition pore inhibitors should be tested.

References

Gryshchenko O, Gerasimenko JV, Peng S, Gerasimenko OV & Petersen OH (2018). Calcium signalling in the acinar environment of the exocrine pancreas: physiology and pathophysiology. *J Physiol* **596**, 2663–2678.

Hegyi P & Petersen OH (2013). The exocrine pancreas: the acinar-ductal tango in physiology and pathophysiology. *Rev Physiol Biochem Pharmacol* **165**, 1–30.

Hegyi P & Rakonczay Z Jr (2011). The role of nitric oxide in the physiology and pathophysiology of the exocrine pancreas. *Antioxid Redox Signal* **15**, 2723–2741.

Jakubowska MA, Ferdek PE, Gerasimenko OV, Gerasimenko JV & Petersen OH (2016). Nitric oxide signals are interlinked with calcium signals in normal pancreatic stellate cells upon oxidative stress and inflammation. *Open Biol* **6**, 160149.

Pallagi P, Venglovecz V, Rakonczay Z Jr, Borka K, Korompay A, Ozsvári B, Judák L, Sahin-Tóth M, Geisz A, Schnúr A, Maléth J, Takács T, Gray MA, Argent BE, Mayerle J, Lerch MM, Wittmann T & Hegyi P (2011). Trypsin reduces pancreatic ductal bicarbonate secretion by inhibiting CFTR Cl^- channels and luminal anion exchangers. *Gastroenterology* **6**, 2228–2239.

Additional information

Competing interests

None declared.