

## COMPLEMENT RECEPTOR 3 PLAYS A SIGNIFICANT ROLE IN B-GLUCAN INDUCED ROS PRODUCTION IN PORCINE NEUTROPHILS.

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Pro-and prebiotics are known to stimulate the intestinal immune system. One of the most commonly used immunostimulants in animal husbandry are β-glucans. β-glucans are conserved glucose polymers found in the cell walls of plants, fungi, yeasts and bacteria. Through binding to their receptors they can activate innate immunity, thereby enhancing defense barriers. Several studies in mice have established that dectin-1 is the most efficient receptor on macrophages and dendritic cells for phagocytosis of β-glucan-rich cell walls. In humans, however, a recent study showed an indispensable role for complement receptor 3 (CR3) in phagocytosis as well as 'reactive oxygen species' (ROS) production by neutrophils in response to zymosan (β-glucan derived from Saccharomyces cerevisiae), as blocking antibodies against CR3 completely inhibited the response of neutrophils towards zymosan. As CR3 is also expressed on porcine neutrophils and monocytes, it could be possible that this receptor is critically involved in the response of porcine innate immune cells to β-glucans. To investigate this hypothesis, we studied the antimicrobial activity of porcine neutrophils after stimulation with three different particulate β-glucans (Zymosan, Macrogard and Euglena gracilis). First, the β-glucan receptors dectin-1 and CR3 were blocked. Laminarin was used to inhibit dectin-1mediated responses. The β-subunit of CR3 was inhibited by monoclonal antibodies against CD18 (anti-CD18) and the α-subunit with both anti-CD11R1 and anti-CD11R3. Subsequently, the β-glucans were added to the neutrophils and the ROS production was measured and compared to the noninhibited cells. The results show a significant role of CR3 in pigs, as blocking the α-subunit of this receptor results in a significant reduced inhibition of ROS production by neutrophils. In addition, the antimicrobial activity of porcine neutrophils to β-glucans was only slightly affected by blocking dectin-1 with laminarin. Although dectin-1 was described as the most important β-glucan receptor in mice, we conclude that as for human, CR3 also plays a cardinal role in β-glucan induced ROS production by porcine neutrophils.