Proceedings of the Joint Meeting AGI-SIBV-SIGA
Assisi, Italy – 19/22 September, 2011
ISBN **978-88-904570-2-9**

Poster Communication Abstract – 9.34

OZONE EFFECT ON RAGWEED POLLEN VIABILITY AND NAD(P)H OXIDASE ACTIVITY

PASQUALINI S., EDERLI L.

Department of Applied Biology, University of Perugia, Borgo XX Giugno 74, 06100 Perugia (Italy)

NAD(P)H oxidase, ozone, pollen, ragweed, ROS

Background

The increase in respiratory diseases arising from allergies in industrialised countries in recent years is considered to be linked to changes in certain environmental factors. One such factor relates to the higher levels of atmospheric pollution and the greater presence and distribution of allergenic *taxa.* We investigated the effects of ozone (O_3), the main component of the photochemical smog, by exposing ragweed (*Ambrosia artemisiifolia*) pollen to a high O_3 concentration (100 nL L^{-1}) for 7 days. We specifically evaluated: pollen reactive oxygen species (ROS) and nitric oxide (NO) content, as NO is considered to be a mediator of inflammatory responses; activity of the nicotinamide adenine dinucleotide phosphate (NAD[P]H) oxidase, which can generate ROS; expression of the major ragweed pollen allergens; and pollen viability.

Results

We investigated the impact of O_3 on ROS and allergen content of ragweed pollen. Pollen was exposed to acute O_3 fumigation, with analysis of pollen viability, ROS and NO content, activity of NAD(P)H oxidase, and expression of major allergens. There was decreased pollen viability after O_3 fumigation, which indicates damage to the pollen membrane system, although the ROS and NO contents were not changed or were only slightly induced, respectively. Ozone exposure induced a significant enhancement of the ROS-generating enzyme NAD(P)H oxidase. The major allergenic proteins released from ragweed pollen ranged from 8 kDa to 43 kDa, with similar protein pattern profiles in both control and O_3-fumigated pollen. The major antigenic component of ragweed pollen, Amb a 1, was identified as a band visible at about 38 kDa, and Western blotting analysis revealed that its content did not change after O_3 exposure. We also examined the expression profiles of the major ragweed pollen allergens: Amb a 1 and Amb a 2, which are proteins belonging to the pectate lyase family, and profilin 1 and profilin 2, which are proteins involved in signal transduction from the outer cell membrane to the inside of the cell, and in the regulation of actin polymerisation. Our data show that O_3 exposure did not affect the expression of the major ragweed pollen allergens.

Conclusions

Pollen represents a critical stage in the life cycle of plants, as viable pollen is crucial for efficient sexual reproduction in plants. Our data indicate that exposition of ragweed pollen to realistic O_3 concentrations reduces pollen viability. As the pollen viability is significantly related to pollen germination and the length of the pollen tubes, effects on the reproduction of ragweed in polluted situations should be taken into account. The present study also indicates that there is an impact of the air pollutant O_3 on the ROS/NO content, NAD(P)H oxidase activity and allergens of ragweed pollen grains. While the intrinsic ROS and allergen content were not affected by O_3 fumigation, there was significant enhancement of the activity of the ROS-generating enzyme NAD(P)H oxidase. This enzyme was almost completely released after short times of *in-vitro* pollen

hydration, as occurs in nature when pollen comes into contact with the cells of the respiratory apparatus. We conclude that realistic doses of O_3 can increase ragweed pollen allergenicity through stimulation of NAD(P)H-oxidase-mediated ROS generation at the airways level.