Canine distemper virus (CDV) is a highly infectious and an acknowledged lethal pathogen of many carnivores. The disease is controlled by vaccination, but an increasing number of distemper cases is recorded also in vaccinated dogs all over the world. No therapy is currently available for the treatment of CDV. The only active compound against CDV on the market is Ribavirin, which showed to be highly toxic. Proanthocyanidin A2 (PA2) belongs to flavonoids being a phenolic polymer widely available in plants and fruits with already known antioxidant and antibacterial activity. Flavonoids have been already investigated for their antiviral efficacy and it has been demonstrated a relationship between their structure and the inhibitory activity against RNA viruses. In this study, we evaluated the in vitro antiviral activity against CDV of Proanthocyanidin A2 (PA2). Our results show that PA2 exerts in vitro antiviral activity against CDV with a higher Selectivity Index compared to Ribavirin. To investigate the PA2 mechanism of action, a Time of Addition assay was carried out adding 3 different concentrations (150, 100 and 75 µg/ml) of PA2 at several times post-infection (p.i.). The viral load of supernatants and monolayer cells, collected at different time points, have been assessed by titration and Real Time PCR. The resulted data were correlated to the kinetic of CDV replication. PA2 decreased the viral growth significantly (P<0.05) up to 16 hs p.i. in a time- and concentration-dependent way both in monolayers and insupernatants. CDV RNA-dependent RNA-polymerase complex is mostly expressed up to 20 hs of the replicative cycle and by Real Time PCR we demonstrated that this complex is significantly less expressed in PA2 treated CDV infected cells by quantifying N and P genes expression. These results show that PA2 interferes with CDV RNA synthesis and that the molecular target of PA2 in CDV infection is the RNA replicative complex.

Keywords: canine distemper virus, proanthocyanidin A2, RNA replicative complex