Assessment of Anti-allergic Properties of Proline Rich Polypeptide Extract (PRP) on Systemic Anaphylaxis in Guinea Pigs

INTRODUCTION
Proline rich polypeptides are active immune modulating peptides from colostrum. Specifically, they are thought to help modulate Th1/Th2, favoring a “shift to the left”, meaning they tend to up-regulate Th1 and down-regulate Th2. In order to test PRP’s ability to down regulate Th2, a study was designed to measure PRPs effect the allergic reactions of guinea pigs sensitized to egg protein. Simultaneously, another group of sensitized guinea pigs were used to see the direct antihistamine effect on PRP, if any, upon their exposure to histamine.

MATERIAL AND METHODS
This study was conducted on both male and female guinea pigs. Egg white protein (ovalbumin, GIII Sigma) and histamine (histamine dihydrochloride, Sigma) were used as antigens. Ovalbumin and histamine were injected into animals by using compressor nebulizer (Parl).

P. Anderson Method (1980) of active sensitisation of guinea pigs
Guinea pigs (250-300 g) were sensitized by one time injection of 10 gm ovalbumin (OA) and 100 mg aluminum hydroxide (Al(OH)3) in 1ml total volume: 0.2 ml intramuscularly in thigh on two sides and 0.6ml intra-abdominally. Experiment began 5 weeks after sensitization of exposure to ovalbumin. These findings support their purported function of down-regulating Th2.

EXPERIMENTAL DESIGN
1. Measurements of bronchial spastic duration time after histamine introduction - internal control.
3. Measurement of bronchial spasm after exposure to histamine (first group) and ovalbumin (second group) preceded 30 minutes by oral introduction of exposure to PRP by spray.

Histamine Model: induced bronchial spasm in guinea pigs via histamine. 0.2% histamine solution containing 0.9% of NaCl was inhaled by guinea pigs, which had been kept in special chamber, until the first signs of bronchial spastic reaction. Duration time of this reaction was measured. Development of bronchial spastic reaction is characterized by 2 phases: 1) acute phase – guinea pigs lie on their side, deep breath, frequency of breathing 10-15 per minute; 2) subacute phase – sitting position, increase of chest muscular activity, more frequent breathing 40-50 per minute. Introduction of PRP was done by spray 5 or 10 times into animal mouth-throat 30 minutes before histamine introduction.

Antigen Model: induced bronchial spasm mode in immunized guinea pigs via ovalbumin.
Experiment was conducted on guinea pigs actively sensitized by ovalbumin in accordance with P. Anderson method. Ovalbumin was used as inductor of bronchial spastic reaction. Animals were dived into two groups. Ovalbumin in dose of 2.5 g/kg was given to the animals of first group. Ovalbumin, dissolved in 1 ml of 0.9 % NaCl solution, was inhaled during 3 min with nebulizer followed by assessment of acute and sub-acute bronchial spasm. Animals in the second group received PRP 30 min prior to similar ovalbumin injection. PRP was delivered by the spray into mouth-throat in the amount of 10-12 sprays. Antiallergic effect was evaluated on the basis of difference in extent of acute and sub-acute bronchial spasms between experimental and control groups.

EXPERIMENTAL RESULTS
PRP in the dosages of 5 and 10 sprays didn’t affect the development of bronchial spasms induced by histamine inhalation in guinea pigs. The several minutes of histamine induced acute and subacute phases of bronchial spastic reaction experienced by all the test subjects were practically the same in the control and experimental groups. There were no deaths in either group.

However, in the case of ovalbumin induced bronchial spasms the antiallergenic effect of PRP was significant. Delivery of a permissive dose of ovalbumin resulted in development of bronchial spastic reaction in 100% of animals (6 out of 6), 3 (50%) of whom died from suffocation. The duration of acute phase in the surviving animals (3) was 313 seconds in average; the sub-acute phase averaged 547 seconds. Introduction of PRP prior to ovalbumin inhalation resulted in death of 2 animals out 7 (28.6%).

CONCLUSIONS
- PRP didn’t affect the development of systemic anaphylaxis induced in sensitized guinea pigs by exogen histamine. Proline rich polypeptides do not appear to act as antihistamines
- PRP has shown clear anti-allergic activity and inhibited the development of systemic anaphylaxis, induced in sensitized guinea pigs by ovalbumin. These findings support their purported function of down-regulating Th2 mediated allergic cascades.

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Death Rate in Sensitized Guinea Pigs from Allergy Induced “Asthmatic” Bronchial Spasm Pre PRP and Post PRP*