G. Liccardi[1], M.B. Bilò[2], F. Manzi[1], A. Piccolo[1], E. Di Maro[1], A. Salzillo[1]

What could be the role of molecular-based allergy diagnostics in detecting the risk of developing allergic sensitization to furry animals?

[1]Department of Pulmonology, Haematology and Oncology, Division of Pneumology and Allergology, High Speciality "A.Cardarelli" Hospital, Naples, Italy
[2]Allergy Unit, Department of Immunology, Allergy and Respiratory Diseases, University Hospital Ancona, Italy

Key words
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Corresponding author
Gennaro Liccardi
Department of Pulmonology, Haematology and Oncology
Division of Pneumology and Allergology
High Speciality "A. Cardarelli" Hospital
Piazzetta Arenella 7, 80128 Naples, Italy
Phone: +39 081 747 333 5-4-3
Fax: + 39 081 747 333 1
E-mail: gennaro.liccardi@tin.it

Summary
Although this highly refined diagnostic approach has been used in several fields of allergy diagnosis, we noticed the scarcity of data on the role of CDR in detecting current sensitization to the allergens of common pets (cat / dog) and, especially, its potential usefulness in predicting the risk of sensitization to other furry animals.
Reported data suggest that cross-reacting mechanisms might play an important role in a significant proportion of allergic sensitizations to furry animals (common pets and unusual / exotic mammals) especially in the absence of any possible direct / indirect contact.
In this context an evaluation of specific IgE by using the micro-array technique ImmunoCAP ISAC (Thermofisher Scientific - Immuno-Diagnostics, Sweden) for lipocalins (Can f 1, Can f 2, Equ c 1, Fel d 4, Mus m 1) and albumins (Bos d 6, Can f 3, Equ c 3, Fel d 2) might be very useful to evaluate the possibility of cross-reactions between the allergens of different animals. In fact, allergic sensitization without animal exposure is a relevant risk for patients, because they are not aware about the possibility that even severe respiratory symptoms may develop after an occasional animal contact. This aspect should be taken into account by susceptible individuals before acquiring new pets, after removal of common pets or beginning a contact for working / leisure activity with a common as well as uncommon animal.

Summary statement

Component Resolved Diagnostic (CRD) could be useful in detecting the risk of developing allergic sensitization to furry animals.

To the Editor

Component Resolved Diagnostic (CRD) allows to map allergic sensitization of patients at molecular level by using purified natural or recombinant allergenic molecules (1-4). Although this highly refined diagnostic approach has been used in several fields of allergy diagnosis, we noticed the scarcity of data on the role of CDR in detecting current sensitization to the allergens of common pets (cat / dog) and, especially, its potential usefulness in predicting the risk of sensitization to other furry animals.
Exposure to animal allergens constitutes a relevant risk factor for the development of allergic sensitization and respiratory allergic diseases such as asthma and rhino-conjunctivitis in susceptible individuals (5).
The frequency of ownership and the prevalence of allergic sensitization to cats / dogs varies in different countries according to cultural differences and environmental factors. Their values are particularly high in some Northern European countries (e.g. Denmark and Finland) (6) and in the US (7).
In all industrialized countries, more and more people become owners of less common small mammals as pets (8-10) or are in contact with bigger animals for work or leisure. Although

many allergens from these animals have been identified, several problems still exist on epidemiology, characteristics of exposure and sensitization to these allergens / animals. For example, it has been widely recognized that cat and dog allergens should be considered as ubiquitous since they are found not only in indoor environments containing these animals but also in other indoor private / public places where cats / dogs have been never kept (11)

Accumulation of pet allergens in indoor environments without animals has been demonstrated to correlate with the number of visitors owning a pet or with those who are in regular contact with these animals. Therefore, the higher the pet ownership in a given community, the higher the presence of pet allergens in apparently pet-free spaces (12). In westernized countries, the consequence of pet allergen ubiquity is a persistent stimulation of airways similar to that induced by dust mite. This indirect modality of exposure is likely to be involved also for other animals (13,14).

Another important aspect is that allergic sensitization to furry animals can be induced not only by direct / indirect exposure but also by a cross-reaction mechanism involving some families of allergenic proteins. (15,16).

Lipocalins constitute the most important group of mammalian inhalant allergens because they are the major allergenic materials derived from dog (Can f 1-2), cattle (Bos d 2), horse (Equ c 1), rat (Rat n 1), mouse (Mus m 1), guinea pig (Cav p 1), rabbit (Ory c 1), hamster (Pho s 21) (3). The role of lipocalins is to carry small hydrophobic molecules and pherormones. Some lipocalins show a very low amino-acid identity whereas others greater homologies and IgE cross-reactivity (between 47-67%) such as Fel d 4, Can f 6, Equ c 1, Ory c 4, Mus m 1 and Rat n 1 (17).

Serum albumins represent the major component of proteins in the circulatory system of mammals, their functional role is the control of colloid osmotic blood pressure and the transport of ligands. The molecular weight of serum albumins is 66-69 kDa. It has been shown that mammalian serum albumins exhibit a very high amino-acid identity to human serum albumins (about 72-82%) (18).

Although their diffusion and cross-reactivity are high in mammals, the role of serum albumins in clinical practice is relatively low. Some case reports have shown that albumins are involved in anaphylactic reactions after artificial insemination (19), episodes of food allergy (20) and asthmatic reactions (21,22).

The most important animal allergen of **secretoglobin** family is Fel d 1, the main cat allergen. Recently, Hilger et al. described a second allergen, a rabbit lipophilin Ory c 3 which shares a 24% of sequence identidy with Fel d 1 (23). This means that secretoglobins are not involved in any significant cross-reaction with other described animal allergen families.

We investigated the role of distinct modalities of exposure to animals (direct, indirect, no contact) in sensitized individuals living in urban areas of Naples and Italy and non-occupationally exposed to any animal. Urban area represents a good model to study all possible modalities of exposure to different animals because the population is not extensively exposed.

In Naples area, only about fifty percent of atopic patients sensitized to common pets (cats / dogs) are directly exposed to these animals, whereas the other half are indirectly exposed or not exposed (24). If we consider allergic sensitization and modalities of exposure to other furry animals such as rabbits, hamsters, rats, horses, guinea pigs, cows and mouse the percentage of sensitized individuals exposed directly to these animals ranges between 0-33.3%, whereas patients sensitized to the same animals with indirect or no contact ranges between 66.7-100% (24). These last percentages regard some animals with unlikely "indirect exposure" in urban areas such as horses / cows or animals rarely kept as pets (hamsters and guinea pigs). A cross-reacting mechanism between lipocalins is the only plausible explanation for these allergic sensitizations. High percentages of allergic sensitization to rabbits and horses without any known direct / indirect contact with these animals have been also shown in two Italian multicenter studies (25,26).

Finally, we have recently shown that exposure and allergic sensitization to common pets (cats / dogs) increases by about fourteen-fold the risk of developing sensitization to other furry animals (rabbits, hamsters, rats, horses, guinea pigs, cows and mouse) suggesting a possible predisposition to develop multiple sensitization to animal allergens (allergic phenotype?) (27).

Reported data suggest that cross-reacting mechanisms might play an important role in a significant proportion of allergic sensitizations to furry animals (common pets and unusual / exotic mammals) especially in the absence of any possible direct / indirect contact. In our opinion, an important question is the risk of developing allergic sensitization to "other" furry animals in individuals already sensitized to common pets (cat / dog). If we consider the aforementioned animal allergen families, the risk of becoming sensitized to less common pets or animals is likely much higher in dog in comparison to cat-sensitized individuals, because of the common presence of lipocalins in the most of allergenic materials (**figure 1**). In this context, an evaluation of specific IgE by using the micro-array technique ImmunoCAP ISAC (Thermofisher Scientific - Immuno-Diagnostics, Sweden) for lipocalins (Can f 1, Can f 2, Equ c 1, Fel d 4, Mus m 1) and albumins (Bos d 6, Can f 3, Equ c 3, Fel d 2) might be very useful to evaluate the possibility of cross-reactions between the allergens of different animals. In fact, allergic sensitization without animal exposure is a relevant risk for patients because they are not aware about the possibility that even severe respiratory symptoms may develop after an occasional animal contact (28-

Figure 1 *- Possible risk of developing allergic sensitization to furry animals in individuals already sensitized to cat / dog.*

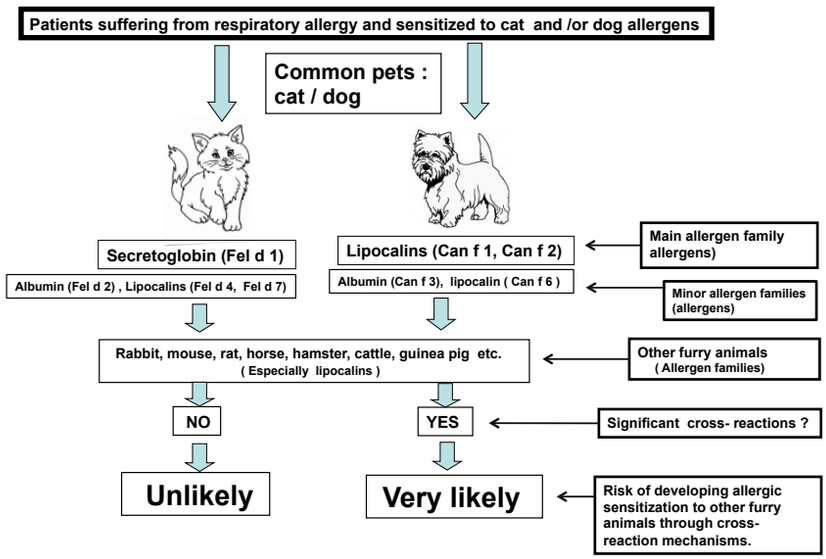
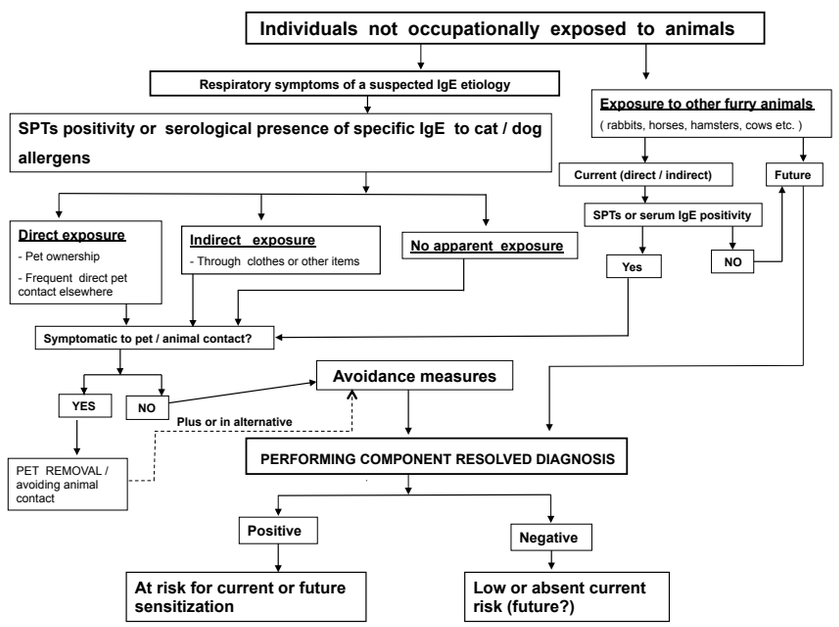


Figure 2 *- Suggested diagnostic flow-chart.*



30). This aspect should be taken into account by susceptible individuals before acquiring new pets after removal of common pets or beginning a contact for working / leisure activity with a common as well as uncommon animal.

In **figure 2** we suggest a possible flow chart in which CRD could be useful to discriminate individuals with a "selective" allergic sensitization (e.g to cat) and showing no IgE production against albumins and lipocalins from patients producing IgE to a wide spectrum of mammal allergens. These last individuals are likely at higher risk of developing future sensitization if directly exposed, for whatever reasons, to furry animals. Recently, we have confirmed these hypotheses by using an *in vitro* model (the micro-array technique ImmunoCAP ISAC, Thermofisher Scientific - Immuno-Diagnostics, Sweden, in 741 subjects referred to the Allergy Unit of Fondazione Salvatore Maugeri, Pavia). These *in vitro* data suggest that allergic sensitization to common pets increases the risk of sensitization to horse and mouse because of the presence of lipocalins. Since lipocalins show a certain degree of cross-reactivity, a similar finding for other furry animals is likely (31).

In conclusion, although CRD has been less studied / used in diagnosis of animal allergy in comparison to other topics (e.g. food / pollen allergy) we think that this diagnostic approach could be very useful in many human / clinical situations if we consider the worldwide interest in owning / working / having leisure with common / less common furry animals.

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