

Letter to the Editor

Majority of shrimp-allergic patients are allergic to mealworm

To the Editor:

The growing world population motivates the exploration of new sustainable protein sources to ensure food security. Insects such as mealworm (*Tenebrio molitor*) are promising candidates, with active ongoing marketing efforts within America and Europe. This warrants assessment of the potential risks. Toxicologic and microbiological risks were assessed previously,^{1,2} but not the potentially allergenic risks. Pilot results³ suggest that shrimp-allergic patients might be at risk for mealworm allergy because IgE binding to tropomyosin and arginine kinase (major shellfish allergens) and sarcoplasmic calcium-binding protein and myosin light chain (minor shellfish allergens) was detected. Our aim was to investigate the allergic potential of mealworm in the shrimp-allergic population.

Fifteen from 60 adult patients from the University Medical Center Utrecht with shrimp allergy based on specialist opinion and diagnostic testing were included in this double-blind, placebo-controlled food challenge (DBPCFC) trial, after testing positive in ImmunoCAP, skin prick test, basophil activation test, or immuno-blot (see this article's [Online Repository at www.jacionline.org](http://www.jacionline.org)).

All subjects gave written informed consent before participation. The study was approved by the local ethics committee (NL43731.041.13).

Patients' median age was 38 years (range, 19-69 years), and 47% were men. Symptoms by history ranged from oral symptoms to anaphylactic shock. Four patients had a positive diagnostic shrimp challenge in the past ([Table I](#)). None of the patients knowingly consumed mealworm proteins. Most had inhalant allergies to house dust mite (11 of 15) and pollen (11 of 15), and 9 patients had 1 or more other food allergies. All patients avoided eating shellfish.

ImmunoCAP to mealworm was positive (>0.35 kU/L) in 9 patients, with a mean of 1.8 kU/L (range, 0.66-6.0 kU/L) (see [Table E1](#) in this article's [Online Repository at www.jacionline.org](http://www.jacionline.org)). The molecular weight of the recognized protein bands ranged from 10 to 200 kDa. Most (14 of 15) recognized tropomyosin and/or arginine kinase (10 of 15). In addition, 4 patients recognized bands with a molecular weight of more than 60 kDa, including a band at 200 kDa, and 4 mainly recognized proteins with a molecular weight of less than 25kDa. Interestingly, IgE from 1 patient bound only to a 200-kDa protein (see [Fig E1](#) in this article's [Online Repository at www.jacionline.org](http://www.jacionline.org)).

Mealworm skin prick test was positive in all patients, and reactivity was comparable to shrimp and HDM. Mealworm basophil activation test was positive for all patients with an approved test (see [Table E2](#) in this article's [Online Repository at www.jacionline.org](http://www.jacionline.org)).

DBPCFC confirmed mealworm allergy in 13 of 15 shrimp-allergic patients ([Table II](#)). Symptoms were oral allergy (7 of 13), urticaria (6 of 13), nausea (8 of 13), abdominal cramping (4 of 13), vomiting (1 of 13), and dyspnea (2 of 13). Subjective symptoms started at a dose of 21.6 mg mealworm protein, and objective symptoms at a 10-fold higher dose (216 mg). One patient had a reaction 4 hours after the final dose, showing both subjective and objective symptoms. Two patients did not complete the DBPCFC because of the severity of their reaction on the first day. Because they both appeared to react to the active dose, it was decided to keep them in the analysis. Severity of reactions to mealworm during DBPCFC was scored between 0 and 3 according to Mueller classification or between 0 and 4 according to Sampson classification ([Table II](#)). The 2 patients with a negative DBPCFC showed no difference in atopic constitution or shrimp allergy with the patients with a positive DBPCFC.

When comparing the mealworm challenge outcome of 4 patients who also had a shrimp challenge, eliciting doses (ED5 and ED10) as well as severity were in the same range. To place

TABLE I. Demographic characteristics, history, SPT, threshold (if performed) to shrimp, and sIgE to relevant components

Patient	Sex	Age (y)	AA/AD/AR	Mueller shrimp history	SPT shrimp	Shrimp CAP (kU/L)	Pen m 1 (ISU)	Pen m 2 (ISU)	Pen m 4 (ISU)	Diagnostic shrimp challenge
1	M	27	N/Y/Y	1	2+	1.3	2.7	0	0	—
2	M	69	Y/Y/Y	1	2+	35	19	0	0	—
3	F	60	Y/Y/Y	1	3+	4.3	0	0.80	0	10 g (2.16 g) obj
4	M	34	N/N/N	2	3+	5.5	12	0	0	—
5	F	46	Y/Y/Y	2	3+	1.9	0	4.2	0	—
6	M	30	Y/Y/Y	3	3+	26	58	0	0	0.3 g (65 mg) subj 1 g (216 mg) obj
7	M	46	Y/N/Y	3	3+	15	18	0	1.2	—
8	F	23	N/N/Y	3	2+	4.6	7.6	1.6	0	0.1 g (21.6 g) subj
9	F	23	Y/Y/Y	3	3+	53	44	33	0	—
10	F	52	N/N/N	4	1+	0.37	0.60	0	0	—
11	M	19	N/N/N	4	1+	6.1	0	0	0	—
12	M	45	N/Y/Y	4	2+	18	0	0	0	—
13	M	26	N/N/N	4	2+	1.9	3.3	0.60	0	1 g (216 mg) obj
14	F	27	Y/Y/Y	4	4+	18	16	4.1	0	—
15	F	47	Y/N/Y	4	1+	2.0	0	0	0	—

SPT in respect to histamine 3+; Pen m 1, shrimp tropomyosin; Pen m 2, shrimp arginine kinase; Pen m 4, shrimp sarcoplasmic calcium-binding protein; Shrimp challenge eliciting dose of shrimp in boldface, protein between parentheses.

AA, Allergic asthma; AD, allergic dermatitis; AR, allergic rhinitis; F, female; ISU, ISAC standardized units; M, male; obj, objective symptoms; SPT, skin prick test; subj, subjective symptoms.

TABLE II. Mealworm DBPCFC in 15 patients

Patient	0.01 g (2.16 mg)	0.1 g (21.6 mg)	1 g (216 mg)	3 g (648 mg)	10 g (2.16 g)	30 g (6.48 g)	60 g (13.0 g)	Mueller mealworm	Sampson mealworm	Diagnostic shrimp challenge
1	—	—	—	—	—	—	—	Negative	Negative	—
2	—	—	—	OA*	OA	—	OA	0	1	—
3	—	—	—	P, U†	—	—	P, U, NC, F	1	2	10 g†
4	—	—	—	—	OA*	OA	OA	0	1	—
5	—	—	—	—	—	—	OA, N, Sw, U†	1	3	—
6	—	—	—	—	—	—	N, AP*	2	2	0.3 g* 1 g†
7	—	OA*	OA	—	OA, AP	OA, AP	ND	2	2	—
8	—	—	—	OA, U†	OA	OA	OA, U, AR, Cj	1	2	0.1 g*
9	—	—	—	—	OA, Dy, NC†	OA, N, S	OA, N, P, Dy	3	4	—
10	—	—	—	—	—	—	—	Negative	Negative	—
11	—	—	—	—	U†	N, AP	ND	2	2	—
12	—	—	—	—	—	N, AP, Cj†	ND	2	2	—
13	—	—	—	—	—	—	U, N†	2	2	1 g†
14	—	—	OA*	—	—	OA	OA, Cj, P, N, V†	2	2	—
15	—	—	U†	U, C	U, C, Dy, W, F, N	ND	ND	3	4	—

Mealworm dose in boldface, amount of protein between parentheses.

AP, Abdominal cramp or pain; AR, allergic rhinitis; C, cough; Cj, conjunctivitis; Dy, dyspnea; F, flushing; N, nausea; NC, nasal congestion; ND, no dose given; OA, oral allergy; P, pruritus; S, sneezing; Sw, difficulty swallowing; U, urticaria; V, vomiting; W, wheezing.

*Symptoms during mealworm challenge: subjective threshold doses.

†Symptoms during mealworm challenge: objective threshold doses.

these eliciting doses in an even broader scope, population threshold dose distribution information for mealworm from all patients was compared with population threshold data for shrimp from the Vital Scientific Expert Panel⁴ (see Table E3 and Figs E2 and E3 in this article's Online Repository at www.jacionline.org). These were comparable.

The most important conclusion from our study is that mealworm allergy is highly likely in shrimp-allergic patients with potentially severe outcome. Most of the shrimp-allergic patients (14 of 15) were sensitized to either mealworm tropomyosin and/or arginine kinase, and 13 of 15 reacted positive in DBPCFC with mealworm, of whom 11 of 15 developed moderate to severe symptoms. IgE binding was found to not only tropomyosin and arginine kinase but also other unidentified proteins. Thresholds for mealworm were comparable to shrimp.

Although the sample size of 15 patients was of limited power, the observed magnitude of effect is nearly fully penetrant for this particular patient group, which is a representation of the shrimp-allergic population in a tertiary center. Although a slight underrepresentation of lower sIgE titers for shrimp was seen in the study population, the statistical and graphical comparison of the severity score of the patient histories showed no clear evidence of deviations from the shrimp-allergic population. The strength of this study was the use of DBPCFC, in addition to a broad panel of *in vitro* and *in vivo* sensitization tests. No other study has assessed allergenicity of insect-based food to this extent.

Finding such a high percentage of shrimp-allergic patients with a probable cross-reactive allergy to mealworm (87%) is striking. One open challenge study with 2 shrimp species (*Penaeus monodon* and *Macrobrachium rosenbergii*) found only half of the patients reacting to both species.⁵ There are no previous DBPCFC studies on cross-reactivity/cosensitization of shrimp-allergic patients with arthropods, but cross-reactivity within crustacean appeared to be as high as 75% between different species, based on sequence homology and serology.⁶ Our data indicate a high degree of cross-reaction/cosensitization even between less related species.

The severity of mealworm allergy varied between mild (oral allergy) and moderate (urticaria and gastrointestinal symptoms) to severe (dyspnea). Five of the patients showed oral or skin/mucosal symptoms (Mueller score 0-1). An open shrimp challenge study reported 80% oral or skin/mucosal symptoms.⁷ A similar profile was found in a DBPCFC study with shrimp in which the majority 70% experienced oral or skin/mucosal symptoms with Mueller score 0 to 1.⁸ Together, these data indicate that mealworm is at least as allergenic as shrimp. Furthermore, it must be stressed that symptoms during DBPCFC are always an underestimation of severity because the reaction is treated when stopping criteria are met.

Comparison of the preliminary population ED10 estimate for mealworm from our study with the ED10 value for shrimp from recent population threshold data from the EuroPrevall project⁸ and the Vital Scientific Expert Panel⁴ indicates a comparable potency for effect elicitation for mealworm and shrimp protein. More mealworm challenge data are needed to confirm this initial analysis. These findings are highly relevant for consumption of insect-containing foods because the individual thresholds for objective symptoms (216 mg of mealworm) are equal to or lower than the amount of mealworm protein that is currently being used in insect snacks. For instance, in the Belgian brand Damhert, which is already on the market in Belgium and the Netherlands, 259 mg of mealworm protein is used in a 20-g snack, where a serving is about 3 of these snacks.

In conclusion, shrimp-allergic patients are at risk when eating mealworm as an alternate source of dietary protein. Because mealworm-containing products are increasingly being consumed in America and European countries, it might be prudent to consider notifying shrimp-allergic patients about this risk.

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Henrike Broekman, MD^{a,*}
Kitty C. Verhoeckx, PhD^{a,b,*}
Constance F. den Hartog Jager, BSc^a
Astrid G. Kruizinga, PhD^b
Marieke Pronk-Kleinjan, RN^a
Benjamin C. Remington, PhD^b
Carla A. Bruijnzeel-Koomen, MD, PhD^a
Geert F. Houben, PhD^{a,b}
André C. Knulst, MD, PhD^b

From ^athe Department of Dermatology/Allergology, University Medical Centre Utrecht, Utrecht, and ^bTNO, Zeist, The Netherlands. E-mail: h.c.h.broekman-4@umcutrecht.nl.

*These authors contributed equally to this work.

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REFERENCES

1. Finke MD. Estimate of chitin in raw whole insects. *Zoo Biol* 2007;26:105-15.
2. Klunder HC, Wolkers-Rooijackers J, Korpela JM, Nout MJR. Microbiological aspects of processing and storage of edible insects. *Food Control* 2012;26:628-31.
3. Verhoeckx KC, van Broekhoven S, den Hartog-Jager CF, Gaspari M, de Jong GA, Wichers HJ, et al. House dust mite (Der p 10) and crustacean allergic patients may react to food containing Yellow mealworm proteins. *Food Chem Toxicol* 2014;65:364-73.
4. Taylor SL, Baumert JL, Kruizinga AG, Remington BC, Crevel RW, Brooke-Taylor S, et al. Establishment of Reference Doses for residues of allergenic foods: report of the VITAL Expert Panel. *Food Chem Toxicol* 2014;63:9-17.
5. Jirapongsananuruk O, Sripramong C, Pacharn P, Udompunturak S, Chinratanapisit S, Piboonpocanun S, et al. Specific allergy to *Penaeus monodon* (seawater shrimp) or *Macrobrachium rosenbergii* (freshwater shrimp) in shrimp-allergic children. *Clin Exp Allergy* 2008;38:1038-47.
6. Sicherer SH. Clinical implications of cross-reactive food allergens. *J Allergy Clin Immunol* 2001;108:881-90.
7. Thalayasingam M, Gerez IFA, Yap GC, Llanora GV, Chia IP, Chua L, et al. Clinical and immunochemical profiles of food challenge proven or anaphylactic shrimp allergy in tropical Singapore. *Clin Exp Allergy* 2015;45:687-97.
8. Ballmer-Weber BK, Fernandez-Rivas M, Beyer K, Defernez M, Sperrin M, Mackie AR, et al. How much is too much? Threshold dose distributions for 5 food allergens. *J Allergy Clin Immunol* 2015;135:964-71.

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Supplementary Data:

Study design

Medical histories were obtained and sensitization to food allergens (shrimp, other shellfish, and wheat), inhalant allergens (house dust mite (HDM), cat, dog, birch- and grass pollens) and insect allergens (mealworm, cockroach and silkworm) determined. Patients sensitized to mealworm participated in a double blind placebo controlled food challenge (DBPCFC) with mealworm. Patients were excluded when pregnant, diagnosed with severe asthma (FEV1 <70%), or using systemic immunosuppressants or beta-blockers.

Eligible patients were ranked according to sIgE titers to shrimp and those with the highest titers were invited first, since for further serology, higher titers were preferred. Because most of these invited patients had a pollen allergy, we made sure that subsequently patients without pollen sensitization were invited, to avoid selection bias. Population size analyses using binominal distribution, indicated that 15 patients were needed to be included in the DBPCFC with mealworm, to calculate the possibility of allergy with a statistical confidence of 90%, if 15% of the sensitized patients would react to DBPCFC. This 15% was (based on the 30% prediction value of the SPT from a recent shrimp study (1)) used to prevent too limited power, since sensitization testing methods for mealworm were used without any knowledge on the positive predicting values and due to the absence of experience with the mealworm extracts.

Specific IgE

Specific IgE using ImmunoCAP (HDM, shrimp and mealworm) and ImmunoCAP ISAC was tested according to the manufacturer's recommendations (Thermo Fisher Scientific, Uppsala, Sweden), and

expressed in kU/L and ISU respectively. Tests were considered positive with a value of 0.35 or higher in ImmunoCAP and 0.3 or higher in ImmunoCAP ISAC.

Mealworm extract

Fresh and freeze dried Yellow mealworms in final larval stage, were kindly provided by Dutch insect farm Krecia (Ermelo, the Netherlands). Tris/urea and SDS/DTT extracts were prepared as described previously (2) and were used for immunoblot and BAT. SPT solution of mealworm (0.4 mg/mL), was kindly provided by ALK (ALK-Abelló, Spain).

Immunoblot

For SDS-PAGE, the Criterion system with a 10-20 % Ready Gel® Tris-HCl gel (Bio-Rad, Hercules, CA,USA) was used according to the manufacturer's instructions. Mealworm, shrimp (ALK) and shrimp tropomyosin Pen a 1, (Indoor Biotechnologies) extracts (5 µg) were loaded on the gel, under reducing conditions (Laemmli buffer). After protein separation, immunoblot was performed as described previously (2).

Basophil activation test (BAT)

BAT was performed as described previously with minor modifications. Cells were incubated with a dilution series (1:10⁷ to 1:10²) of mealworm extracts (Tris/urea (5 mg/mL) and SDS/DTT). Shrimp extract (ALK, 2 mg/mL), and shrimp tropomyosin Pen a 1 (Indoor Biotechnologies, 1mg/mL), were used as positive controls (2). The results were expressed as a percentage of CD63+ basophils.

Skin Prick test (SPT)

Commercial SPT solutions for HDM, shrimp, common inhalant allergens and wheat (ALK), crab, lobster, cockroach and silkworm (Greer) were used. SPT was performed according to the procedure described previously (3). SPT reactivity was measured as the ratio of the mean of the wheal elicited by the tested extract and histamine control (ratio 1:1 expressed as 3+). When the ratio was 0.5 or greater, the reaction was regarded as positive.

Food challenge

Blanched mealworm was added to chicken meat with added salt, pepper and 0.3% nutmeg. In the placebo, the mealworm was replaced with chicken meat. Patients received 7 portions on each challenge day, starting at 2.16 mg mealworm protein (corresponding with 10 mg total mealworm), followed by 21.6 mg, 216 mg, 648 mg, 2.16 g, 6.48 g, and 13.0 g mealworm protein.

The challenge was discontinued and considered positive in case of objective symptoms or if a suggestive moderate to severe subjective symptom lasted for > 45 minutes, and scored according to Mueller and Sampson (4,5).

Four patients had an open food challenge to boiled Dutch shrimp, prior to participating in this study, consisting of 7 dosing steps (0.1 g, 0.3 g, 1 g, 3 g, 10 g, 30 g and 100 g shrimp).

Analysis

Log-logistic, log-normal, and Weibull eliciting dose (ED) with ED5, ED10, indicating where 5% and 10% of the allergic population is predicted to react were estimated for mealworm using cumulative dose probability distribution models.

Remarks:

We enrolled 18 patients in this study to include 15 patients in the DBPCFC. Three patients were excluded from further analysis due to an exercise induced allergy in one, one for lack of evidence for mealworm sensitization and in one an inconclusive mealworm food challenge result. Sensitization was 94.4% within the invited shrimp allergic patients. In total, we completed our study with 15 patients.

(Supplementary Table E1, Table E2 and Figure E1)

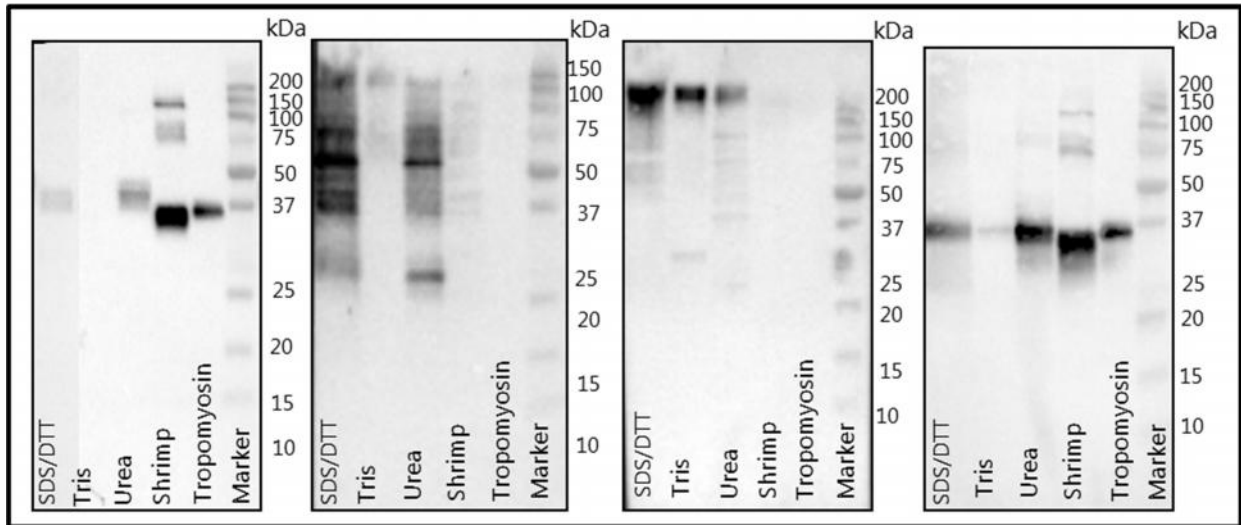
(Supplementary Table E3, Figure E2 and E3)

References used in supplements:

1. Yang AC, Arruda LK, Santos ABR, et al. Measurement of IgE antibodies to shrimp tropomyosin is superior to skin prick testing with commercial extract and measurement of IgE to shrimp for predicting clinically relevant allergic reactions after shrimp ingestion. *J Allergy Clin Immunol*. 2010;125(4):872-8.
2. Broekman H, Knulst A, den Hartog Jager S, et al. Effect of thermal processing on mealworm allergenicity. *Mol Nutr Food Res*. 2015. Epub 2015/06/23.
3. Peeters KA, Nordlee JA, Penninks AH, et al. Lupine allergy: not simply cross-reactivity with peanut or soy. *The Journal of allergy and clinical immunology*. 2007;120(3):647-53. Epub 2007/07/20.
4. Mueller HL, Diagnosis and treatment of insect sensitivity, *The Journal of Asthma Research*, 1966;3(4):331-333.
5. Sampson HA, Anaphylaxis and emergency treatment. *Pediatrics* 2003;111:1601-8.
6. Taylor, S.L., et al., Establishment of Reference Doses for residues of allergenic foods: report of the VITAL Expert Panel. *Food Chem Toxicol*, 2014. 63: p. 9-17
Mueller HL, Diagnosis and treatment of insect sensitivity, *The Journal of Asthma Research*, 1966;3(4):331-333.
7. Ballmer-Weber, B.K., et al., How much is too much? Threshold dose distributions for 5 food allergens. *J Allergy Clin Immunol*, 2015. 135(4): p. 964-71
Sampson HA, Anaphylaxis and emergency treatment. *Pediatrics* 2003;111:1601-8.

Supplementary Material

Figure E1. Representative immunoblots from 4 shrimp allergic patients



Immunoblot in 4 shrimp allergic patients using mealworm extracts in tris, urea and SDS/DTT buffer, shrimp (crangon crangon from ALK) and shrimp tropomyosin (Pen a 1 from indoor technologies), showing varying patterns.

Figure E2. Log-logistic, log-normal, or Weibull probability distribution models of mealworm (expressed as cumulative mg mealworm protein). Predicted distributions and actual challenge data points (•) are displayed.

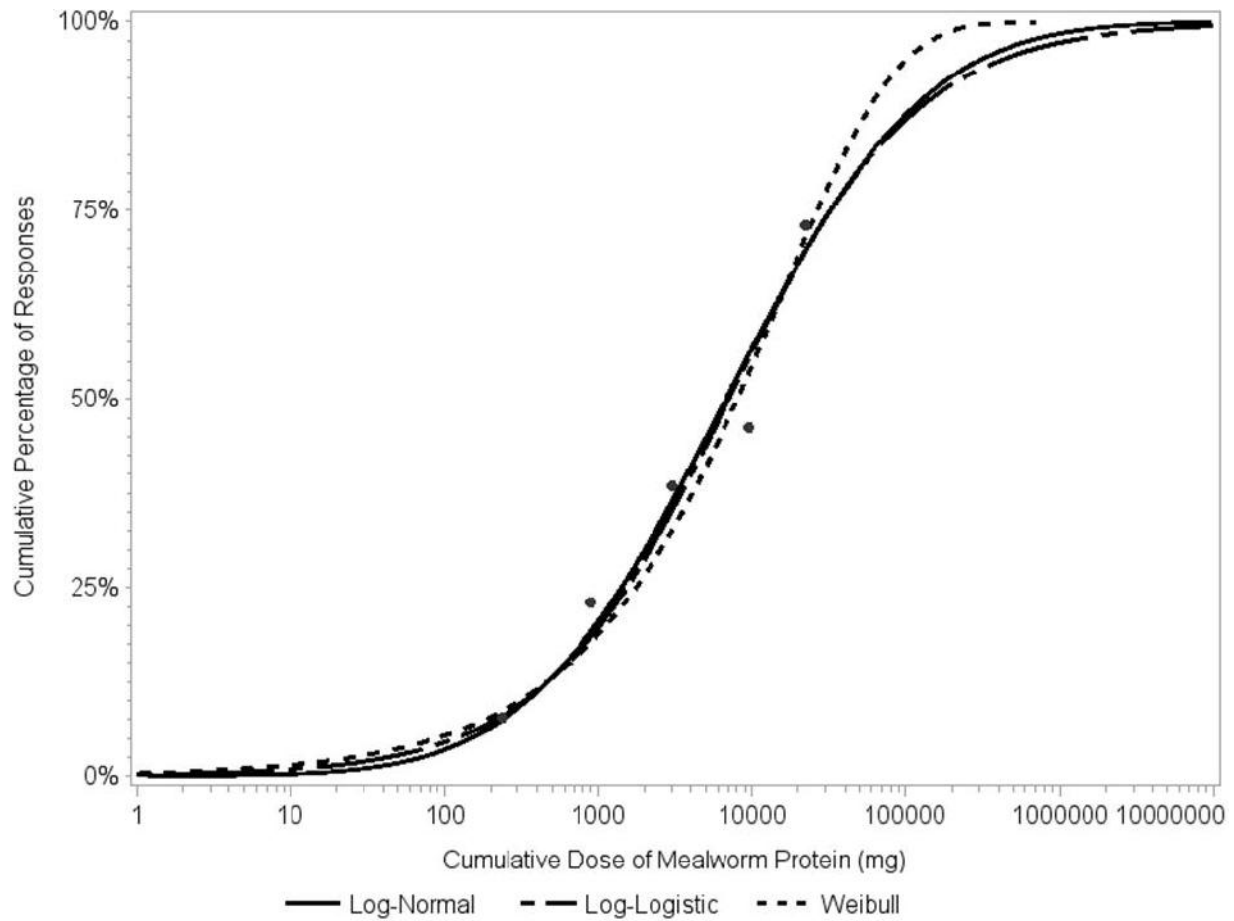
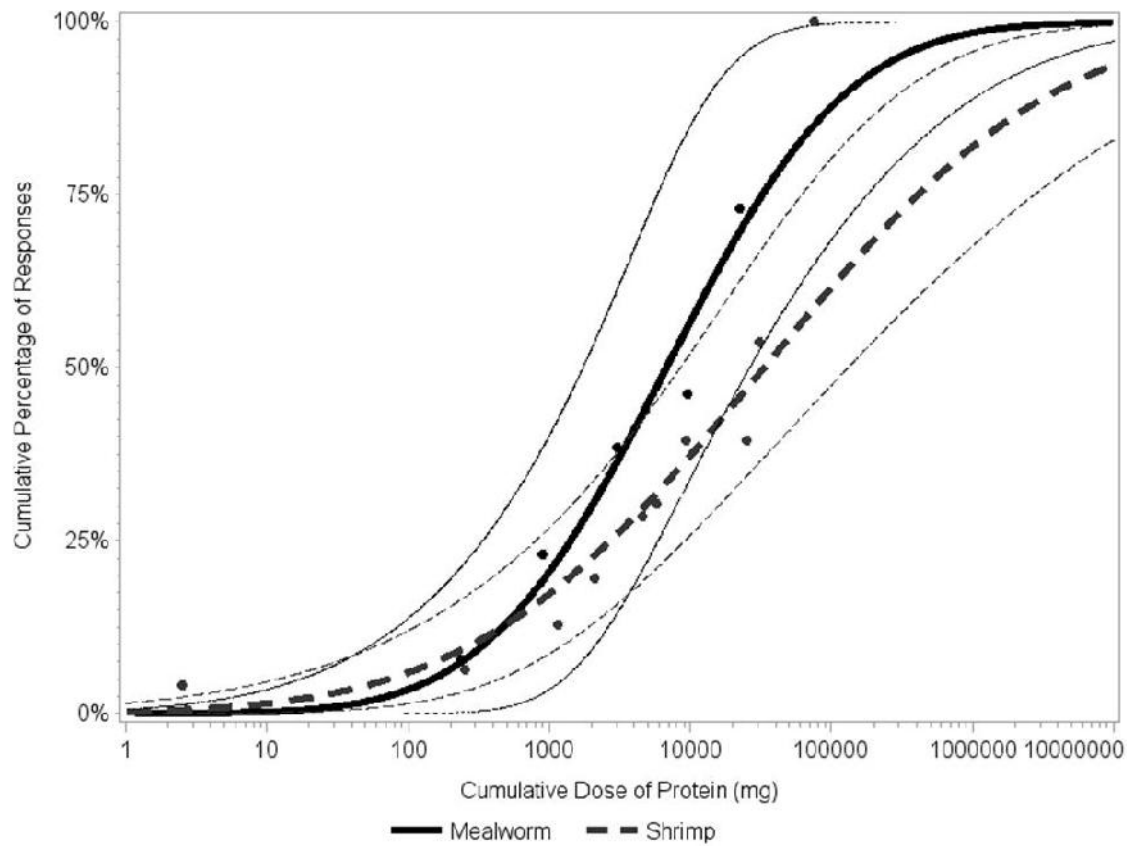


Figure E3. Log-normal probability distribution models of mealworm and shrimp (expressed as cumulative mg protein) and corresponding 95% confidence intervals.



Population threshold data for shrimp analyzed by the Vital Scientific Expert Panel (VSEP) ⁶ and the EuroPrevall project ⁷.

Table E1. Sensitization to tropomyosin, crustaceans, cockroach and silkworm by ImmunoCAP ISAC and/or SPT.

Pt.	Der p 10 (ISU)	Ani s 3 (ISU)	Bla g 7 (ISU)	SPT crab	SPT lobster	SPT cockroach	SPT silkworm
1	3.6	1.6	2.2	2+	2+	2+	1+
2	12	20	14	nt	nt	nt	nt
3	0	0	0	2+	2+	2+	2+
4	9.3	13	11	3+	3+	2+	0
5	0	0	0	nt	nt	nt	nt
6	53	41	44	2+	2+	2+	2+
7	16	7.4	19	3+	3+	3+	2+
8	4.7	6.5	7.5	2+	2+	2+	2+
9	50	40	46	nt	nt	nt	nt
10	0.90	0.60	0.60	1+	0	2+	1+
11	0	0	0	2+	2+	3+	2+
12	0	0	0	2+	2+	3+	3+
13	2.7	4.7	2.7	2+	3+	2+	2+
14	20	17	15	4+	4+	3+	2+
15	0	0	0	1+	2+	2+	2+

Der p 10 house dust mite tropomyosin; Ani s 3 anisakis tropomyosin; Bla g 7 cockroach tropomyosin; ISU ISAC Standardized Units; nt not tested

Table E2. Sensitization patterns to mealworm (n=15):SPT, ImmunoCAP, BAT and immunoblot.

Pt.	SPT mealworm	Mealworm CAP (kU/l)	CD63 release mealworm	CD63 release shrimp	CD63 release Pen a 1	blot trop. mealworm	blot a-k. mealworm	blot shrimp	blot Pen a 1
1	2+	0.16	+	+	+	+	-	+	+
2	2+	3.1	NR	NR	NR	+	+	+	+
3*	2+	0.20	+	-	+	+	+	+	-
4	2+	1.2	+	+	+	+	+	+	+
5	3+	0.31	+	+	+	+	+	+	+
6	2+	4.0	SR	SR	SR	+	+	+	+
7	2+	2.5	+	+	+	+	-	+	+
8	2+	0.80	NR	NR	NR	+	-	+	-
9	4+	6.0	+	+	+	+	+	+	+
10	1+	0.07	NR	NR	NR	+	+	+	+
11	2+	0.30	+	+	+	+	-	+	-
12	2+	1.6	+	+	-	-	-	+	-
13	2+	0.21	+	+	+	+	+	+	+
14	4+	0.66	+	+	+	+	+	+	+
15	3+	2.3	+	+	+	+	+	+	-

Pen a 1 = shrimp tropomyosin NR = non-responder (control negative) SR = spontaneous release (spontaneous release on negative control) s.trop = shrimp tropomyosin pen a 1, trop = tropomyosin, a-k = arginine kinase

*suspected switch in BAT assay shrimp and tropomyosin results.

Supplementary Material Table 3.

Table E3. Log-logistic, log-normal, and Weibull ED estimates from the cumulative dose mealworm probability distribution models.

	ED05 (95% CI)	ED10 (95% CI)	ED50 (95% CI)
Log-logistic	118 (9.2, 1522)	334 (41.4, 2693)	7058 (1808, 27550)
Log-normal	149 (17.5, 1270)	346 (55.9, 2145)	6754 (1735, 26296)
Weibull	89.2 (4.1, 1954)	309 (27.3, 3513)	8032 (2511, 25696)

CI confidence intervals expressed as mg mealworm protein