

热加工与非热加工技术对水产品致敏性的影响研究进展

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摘要: 我国作为水产养殖和消费总量第一大国, 水产品引发的过敏问题越来越受到关注, 近些年来, 因食用水产品而导致的过敏事件日益增多。该研究综述了近年来热加工技术(蒸、煮、高温压力)、非热加工技术(超高压、低温等离子体、超声波和辐照)以及两种加工技术联用对水产品过敏原消减的研究, 指出热加工技术主要通过使蛋白质变性来消减过敏原的致敏性, 非热加工技术则通过掩盖或破坏过敏原抗原表位来消减过敏原致敏性, 为低致敏性水产品开发提供了重要基础和技术参考。不断探究过敏原诱导过敏反应发生的机理, 加快推进低致敏性水产品加工技术在实际生产中的应用, 有利于控制和降低水产品过敏所带来的风险。

关键词: 水产品; 致敏性; 热加工技术; 非热加工技术

文章编号: 1673-9078(2022)08-327-333

DOI: 10.13982/j.mfst.1673-9078.2022.8.1059

Research Progress on the Effects of Thermal and Non-thermal Processing Technologies on the Allergenicity of Aquatic Products

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Abstract: China is the largest country in aquaculture and total consumption of aquatic products, and the allergic problem caused by aquatic products has attracted more and more attention. In recent years, incidence of allergy caused by the intake of aquatic product has increased gradually. This paper reviews recent research on the allergen reduction of aquatic products by thermal processing technologies (steaming, boiling and high temperature pressure), non-thermal processing technologies (ultra-high pressure, cold plasma, ultrasound and irradiation), and the combination of the two processing technologies. It is pointed out that thermal processing technologies mainly reduce allergenicity *via* denaturing the proteins, whilst non-thermal processing technologies reduce allergenicity *via* masking or destroying the antigenic epitopes of allergens. These results provide an important basis and technical reference for further development of hypoallergenic aquatic products. Continuing investigations on the mechanisms underlying the allergic reactions caused by allergens, and accelerating the development of the application of hypoallergenic aquatic processing technologies in practical production, are both conducive to controlling and reducing the risks associated with aquatic product allergy.

Key words: aquatic products; allergenicity; thermal processing technology; non-thermal processing technology

引文格式:

成军虎,马筱冉,陈璐,等.热加工与非热加工技术对水产品致敏性的影响研究进展[J].现代食品科技,2022,38(8):327-333

CHENG Junhu, MA Xiaoran, CHEN Lu, et al. Research progress on the effects of thermal and non-thermal processing technologies on the allergenicity of aquatic products [J]. Modern Food Science and Technology, 2022, 38(8): 327-333

收稿日期: 2021-09-22

基金项目: 国家自然科学基金面上项目(31972205); 厨房电器健康烹饪技术创新联盟(2021029)

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食物过敏指的是人们对某些食物产生的一种不良反应, 多表现于皮肤或者胃肠道中, 在医学上是一种变态反应, 目前可分为免疫球蛋白E (Immunoglobulin E, IgE) 介导与非 IgE 介导两大类^[1]。调查显示, 水产品、鸡蛋与牛乳是过敏原的主要来源, 有 6% 的年

轻人经历过食物过敏^[2],特别是免疫系统未发育完全的婴幼儿。水产品因其味道鲜美且含有丰富的营养物质而深受人们喜爱,而其引发的过敏问题日渐突出^[3,4],过敏发病率持续上升,鱼、虾等水产品是常见的过敏原^[5]。水产品中的过敏原可以分为鱼类过敏原、甲壳类动物过敏原以及软体动物过敏原。鱼类过敏原包括小清蛋白(Parvalbumin, PV)、鱼卵蛋白(Carp vitellogenin)、胶原蛋白(Collagen),而甲壳类动物和软体动物的过敏原主要是原肌球蛋白(Tropomyosin, TM)^[6]、卵清蛋白(Ovalbumin)和精氨酸激酶(Arginine kinase, AK)^[7]等。水产品过敏会造成机体不适,部分表现为身体某些部位(脸部和腿部等)起疙瘩并伴有瘙痒症状,严重者还会休克甚至威胁生命,水产品致敏性问题亟待解决^[8]。目前除了避免水产品过敏原摄入以外,较为有效地避免水产品过敏的方法为用热加工与非热加工技术处理水产品,热加工与非热加工技术通过修饰或改变过敏原蛋白质的分子结构进而在一定程度上消减过敏原的致敏性,以期作为低致敏性食品的开发提供重要理论基础与技术支撑。

1 水产品过敏原消减技术

1.1 热加工技术

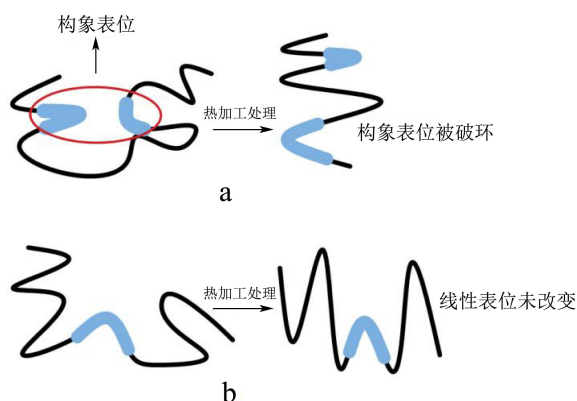


图1 热加工诱导抗原表位变化示意图^[11]

Fig.1 schematic diagram of antigen epitope changes induced by thermal processing

注:(a)热加工诱导的过敏原蛋白质构象表位变化;(b)热加工诱导线性表位的变化。

热加工技术是一类以热能为基础的加工技术,按照加工时是否需要用到水,可粗略地分为湿加热和干加热,前者主要包括煮沸、煎炸、高压灭菌等,后者包括烘烤与微波加热等。这类技术通过将热能逐渐传递或转化给食品而使其组分在热能作用下发生结构变化,从而引起功能性变化^[9]。热加工技术对过敏原影响的作用机制如图1所示,主要是引起过敏原构象表

位变化,对线性表位的作用不明显^[10,11]。

水产品中的TM蛋白是一种热稳定性蛋白,经过热加工(主要是蒸、煮)之后,其二、三级结构会发生变化,从而改变致敏性^[12]。刘思寒等^[13]利用酶联免疫吸附法(ELISA)对凡纳滨对虾主要过敏原TM热加工前后消化产物的IgE结合活性进行分析,发现煮制组虾TM结构的IgE结合活性降低了54.0%±6.1%,而高温压力组虾TM的IgE结合活性降低了64.6%±3.3%,结果表明,蒸、煮处理可显著降低TM蛋白的IgE结合能力,在辅助加压的条件下致敏性可进一步降低。郑礼娜等^[14]进一步对刀额新对虾的虾肉分别进行了煮(100℃)、蒸(100℃)和高温压力(121℃、0.1MPa)处理,利用SDS-PAGE电泳、免疫印迹分析和ELISA对过敏原免疫活性进行定性以及定量检测,发现经过热加工后过敏原免疫活性均有一定程度的降低,但是不同热加工条件的影响程度不同,煮、蒸5min后分子量为36ku的主要过敏原蛋白致敏性分别降低了53%和23%,随着热加工时间的增加,过敏原致敏性呈递减趋势,但变化幅度不显著;而高温压力处理5min后,分子量为36ku的主要过敏原蛋白致敏性降低了78%,30min后,致敏性降低了97%左右,几乎完全消失。由此可见,热加工中高温压力的施加可使虾过敏原致敏性显著下降。

然而,仍有较多研究表明,热加工对水产品过敏原致敏性几乎无作用^[15,16]。Yu等^[17]研究发现,普通蒸煮对拟穴青蟹蟹肉过敏蛋白的致敏性无显著影响,类似地,Usui等^[18]用圆二色谱证明了TM的 α -螺旋结构在80℃热加工之后发生了折叠现象,而恢复到25℃后TM的结构也再次恢复,过敏原致敏性因而没有受到影响。同样,Misnan等^[19]发现蒸、煮处理对当地海螺的蛋白过敏原致敏性影响较小。

此外,还有一些研究表明,热加工可能会使水产品过敏原致敏性增加。Larly等^[20]发现从花尾虾中提取出的过敏原经5到25min的煮沸处理之后,IgE结合活性从0.251nm升到了0.268nm,这说明热加工可能会暴露更多的IgE结合位点。张轶群等^[21]通过对南美白对虾的过敏蛋白与果糖进行混合热加工12h,测得虾蛋白过敏原致敏性增加了44%,可能是加工过程中的美拉德反应使过敏蛋白的三级结构改变,从而导致过敏原抗原表位的暴露,增加了过敏蛋白的致敏性^[22]。

热加工操作简易方便,技术发展较为成熟。根据热加工作用条件的不同,降低水产品致敏性的效果可能会有很大差距。尽管可能产生一些副产物,其仍广泛应用于低致敏性水产品的研发中。在使用热加工技术处理水产品时,应注意热加工的时间、温度以及

辅助加工方式对过敏原致敏性产生的不同程度的影响。

1.2 非热加工技术

非热加工技术是新型的食品加工技术,主要包括超高压、低温等离子体、超声波和辐照等技术。这些技术通过暴露或掩盖食物中过敏原的抗原表位,改变过敏原 α -螺旋和 β -折叠等二级结构,破坏维持三、四级结构的非共价键如氢键、疏水相互作用等方式来改变致敏蛋白的空间结构,从而消减其致敏性^[23]。非热加工技术对食品的固有营养成分影响较小,同时又能很好地维持食品的质构、色泽、新鲜度等,逐渐成为食品加工行业新的推动力^[24-26]。

1.2.1 超高压技术

超高压又称超静压处理技术,是一种新型食品脱敏加工技术,通过破坏蛋白质的非共价键(离子键、氢键和疏水键)和改变蛋白质的高级结构来改变过敏原的致敏性^[27,28]。超高压加工技术对致敏性的作用机制如图2所示,一方面,由于超高压处理后过敏原被溶解、抗原表位被掩盖或破坏,导致蛋白致敏性降低;另一方面,超高压处理可以产生新的抗原表位,从而增强过敏原致敏性^[11]。

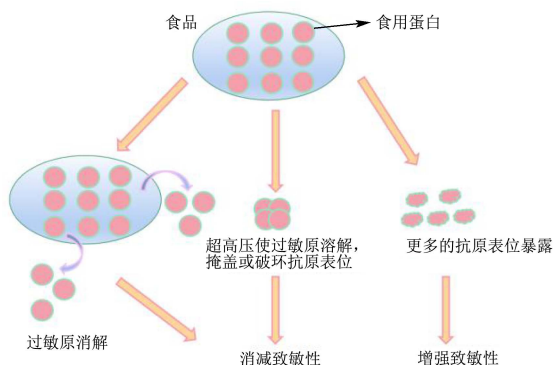


图2 超高压诱导的致敏性变化示意图^[11]

Fig.2 Schematic diagram of ultra-high pressure induced sensitization changes

韩建勋等^[29]研究发现,在100℃下,虾TM蛋白的致敏性随压力的增加而降低,这可能是由于TM结构随压力的增加而不断变化所致。董晓颖等^[30]研究发现,虾过敏蛋白经不同压力处理后,分子量大小虽然没有产生变化,但在100~400 MPa区间,随着压力的增大,虾过敏蛋白的致敏性降低,在400 MPa时降至最低,但在500 MPa时致敏性又开始增加,这可能是由于在100~400 MPa区间,压力的增加导致蛋白质结构发生变化,破坏或掩盖了过敏原表位,从而使其致敏性降低;而在500 MPa时,之前被掩盖的抗原表位会重新暴露,致敏性又会增加。而用200、400和600

MPa的高静压处理鱿鱼中的过敏原TM发现,蛋白质折叠和二级结构改变使得致敏性降低,当压力为400和600 MPa时的致敏消减效果最好^[31]。

除此之外,超高压会导致过敏原蛋白溶出^[32]。牡蛎经过超高压处理后,蛋白和灰分会随汁液向外流出,且牡蛎流失液中游离氨基酸含量随着压力的增大而增大,表明超高压处理可以使某些蛋白水解酶活性增强;同时差式扫描量热(DSC)图谱结果显示,未经处理的牡蛎蛋白有两个吸收峰,且200和400 MPa处理后的吸收峰没有变化,而600 MPa处理后的吸收峰趋于平缓,说明超高压处理使牡蛎的蛋白结构发生变化,过敏原表位遭到破坏,致敏性从而被消减^[33,34]。

1.2.2 低温等离子体技术

低温等离子体(Cold plasma, CP)技术是一种新型食品加工技术。等离子体作为物质的第四态,是由离子、紫外光、电子、自由基和激发原子等活性物质组成的电离气体,这些活性物质可以与蛋白质相互作用并改变其构象^[35]。研究表明,长时间的CP处理导致过敏原蛋白质二级结构中 α -螺旋和 β -折叠含量的改变^[36,37],同时CP可以通过改变过敏原的抗原表位来影响其致敏性,具有降低水产品过敏原致敏性的潜力。Flora-Glad等^[38]用CP处理凡纳滨对虾的TM发现,处理15 min以后, TM结构中的 α -螺旋数量减少, β -折叠增加,表面疏水性和二硫键含量发生显著变化,这些二、三级结构的变化会共同影响TM的致敏性,经ELISA检测证实其IgE结合能力最大下降了17.6%。低温等离子体技术对环境友好,操作简易,消减致敏性的效果明显,但是用于低致敏水产品的开发还需要从加工设备、工艺条件以及安全性等方面进一步研究。

1.2.3 超声波技术

超声波(Ultrasonic wave)是一种频率高于20000 Hz的声波,其方向性好,反射能力强,易于获得较集中的声能。超声能够产生空穴效应、微流束效应以及自由基,并且在超声过程中,超声波在媒质中形成的介质粒子机械振动可产生与媒质间的机械作用、空化作用及热作用,从而使蛋白质结构发生改变^[39,40]。由于超声波技术一般在室温下进行,对环境较友好,作用效率高,可最大程度地避免水产品品质的下降,因此,其在消减过敏原致敏性方面的研究得到了广泛关注^[41,42]。马涛等^[43]研究了超声波处理对三文鱼过敏蛋白致敏性的影响,发现三文鱼致敏蛋白Sal s 1经超声波处理60 min后,过敏原致敏性下降了20.58%。Dong等^[44]在室温下对虾样品进行超声波处理发现,20 min时, TM致敏性的降低效果最好,消减了约76%,而处理时间进一步增加时,观察到多肽的产生和样品碎

片量的增加, 虾的原有口感可能发生了变化, 因此, 在消减水产品致敏性的同时还应考虑水产品本身的性质及品质, 保证其风味、口感、营养成分的最大保留。

1.2.4 辐照技术

辐照技术是利用 γ 射线、电子束射线和X射线对过敏原进行的电离辐射, 使过敏原蛋白发生交联和分子构象的变化, 从而消减过敏原致敏性的技术^[45-47]。

Liu等^[48]对冷冻虾的TM进行3 kGy的电子束辐照后发现, TM的IgE结合活性增加了约10%, 随后把剂量提高至10 kGy后, IgE结合活性下降了20%, 说明随着处理条件的改变, TM致敏性发生了不同的变化。而Guan等^[49]对鲜虾的TM进行剂量为1、3、5、7和9 kGy的电子束辐照后发现, TM的IgE结合能力均显著降低, 剂量为7 kGy时的效果最显著, 结合能力降低了59%, 同时TM的分子量没有显著变化, 这可能是由于辐照导致了TM的结构变化, 如 α -螺旋和 β -折叠的减少和 β -转角的增加, 从而改变了致敏性。Li等^[50]发现, 3~10 kGy的剂量使得TM的结合活性持续增加, 而10 kGy以上的剂量则会使致敏性持续降低。而对于蟹类, 刘光明等^[51]也发现10 kGy以上的剂量处理会使中华绒螯蟹和梭子蟹的TM致敏性大幅降低。同时发现, 10 kGy以上的剂量处理对整蟹纤维肌源蛋白(含TM)的致敏性无显著影响, 可能是蟹肉组织中的其他物质可以保护TM结构不受破坏。由于较大剂量的辐照对食品理化性质的具体作用尚不明确, 为了保留水产品原有的营养和风味物质, 我国农业相关标准中有规定, 应该用4~7 kGy剂量的辐照对冷冻水产品进行处理, 因此, 辐照射线应该在相关标准的剂量规定下使用, 而具体的处理条件控制依据、对致敏原蛋白的影响机理还需要进一步探究。

1.3 热加工与非热加工技术协同

由于单一的热加工技术(蒸、煮)对水产品致敏性的消减效果具有一定的局限性, 比如单一蒸、煮处理对海螺蛋白过敏原的致敏性影响较小^[19], 对拟穴青蟹蟹肉过敏蛋白致敏性消减无显著影响^[17]等。因此, 研究人员开始探索复合加工技术, 以提高过敏原的消减率。Ganesan等^[52]采用蒸煮、超声波-蒸煮、反压-蒸煮和高温高压-蒸煮方式处理蟹肉发现, 蒸煮处理没有显著消减蟹肉TM粗蛋白消化产物的致敏性, 而结合超声波、高温高压和反压处理后, 蟹肉TM的致敏性显著消减, 其中高温高压-蒸煮与反压-蒸煮的消减效果更好。四种加工方式中高温高压-蒸煮与反压-蒸煮消减过敏原致敏性作用效果最好, 超声波-蒸煮次之, 蒸煮效果一般。这些研究也进一步说明不同加工

技术联用可以弥补单一热加工在消减过敏原致敏性方面的不足。

2 总结展望

基于水产品的过敏特性, 本文综述了热加工与非热加工技术对水产品过敏原致敏性的影响, 指出了热加工(蒸、煮)主要通过使蛋白质变性来消减过敏原的致敏性; 非热加工技术主要通过掩盖或破坏过敏原抗原表位来达到消减致敏性的目的, 其良好的技术特性使得其消减过敏原致敏性时能保持食品的风味以及固有特性, 在水产品的脱敏加工中具有一定的潜力。然而, 目前针对部分新型非热加工技术的研究和实践不够充分。对于某些水产品而言, 单一使用某种加工技术的效果不够理想, 因此, 研发低致敏性水产品应该把重点放在寻找并验证降低致敏性效果好、对原料营养成分以及风味损伤最小的加工条件, 而如何将不同技术有效联合以实现最好的过敏原消减效果、建立更多样的过敏原致敏性消减技术是今后的研究方向。

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