Evolution of Acrylamide Detection Methods in Cooked Foods since its firstly Discovered in 2002
Review Article

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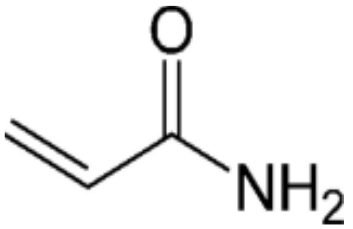
Abstract
The changing of contemporary life with fast style consequently its need for fast ready meals given raise chemical food safety hazards from thermally processed one. Acrylamide, neurotoxin and DNA damage, has been found in different thermally processed foods such as potato chips, biscuits, and bread. Limit literature was done upon its existence in fried meat and/or fish. This paper provides a comprehensive overview of detection of Acrylamide in different foods since the first discovered in April 2002, in fried food by researchers from the Swedish National Food Administration and the University of Stockholm unite. Recent methods based on the nanotechnology as well as molecular biology are presented. The review shows high sensitivity, selectivity, stability, and repeatability methods as liquid chromatography coupled with tandem mass spectrometry. Also, rapid detection methods with merits of simplicity and portability such as computer vision, ELISA, electrochemical biosensing, and fluorescent biosensing. Some methods are expensive although recent methods seem to be cheaper. Meanwhile, suggestions for further research on rapid methods for detecting Acrylamide are also discussed based on types of foods and technical challenges.

Introduction
Acrylamide, (2-propenamide, C3H5NO (71.09 gmol_1, CASNo. 79-06-1)) a well-known human neurotoxic chemical toxin, was detected in fried or baked food.



Acrylamide ($CH_2 = CHCONH_2$)

Acrylamide (AA) is a low-molecular weight vinylic compound. It is a colorless and odorless crystalline substance and it is highly water soluble, easily reactive in air, and rapidly polymerizable. Thermal food processing especially meat and their products can have some accidental changes as the destruction of amino acids and the synthesis of hazards chemicals as heterocyclic amines, acrolein, furan, and acrylamide. Generally, all cooking methods likely to produce acrylamide in foods except boiling (figure 1). The early study by ***Yasuhara et al. (2003)*** suggested that in the thermal processing of lipid-rich food as meat may also play an important role in acrylamide formation. In spite of the fact that high carbohydrate foods have been widely measured for the estimation of AA, preparing high protein foods like meat at high temperature can also be a reason of generating AA, reaching levels up to 300 mg/kg ***(Kaplan et al., 2009; Delgado-Andrade, et al., 2010 and Demirok & Kolsarici, 2014).***

AA is a Maillard reaction product, which forms instinctively as frying or cooking heat treatment imposed in foods ***(Claus et al., 2008)***. One of the major MRP is acrylamide which is developed mostly from free asparagine and reducing sugar throughout cooking foods at high temperatures (above 120ºC). A nearly recent study done by ***Trevisan et al. (2016)*** confirmed temperature as an important factor for the production of acrylamide in grilled meat. They grilled and fried hamburgers to an internal temperature below 90ºC and found that furosine was formed, When the temperature reached 90ºC and 100ºC, furosine content decreased by 36% and fluorescent compounds increased by up to 98%. Baking meat at 300ºC, the most severe heat treatment studied, resulted in the formation of carboxymethyllysine.

According to Food and Agriculture Organization of the United Nations and World Health Organization (FAO/WHO), daily intake of the AA from the food can be in the range of 0.3– 0.8 μg/kg ***(FAO/WHO, 2002)***. The daily intake of the AA from food 1–10 mg/kg) higher than this value can cause toxic effects on humans.

Acrylamide has been proved to cause genotoxicity, muscle weakness, neurotoxicity, sensory loss, and carcinogenicity. Indeed, acrylamide exhibits potential to alky-late nucleophilic center in DNA molecule and initiates mutations and alternations in gene expression in human cells ***(Oracz, et al., 2011)***. Therefore, this article through the light on the common methods for detection AA as hazards chemical toxin and their ability to application in fried meat.

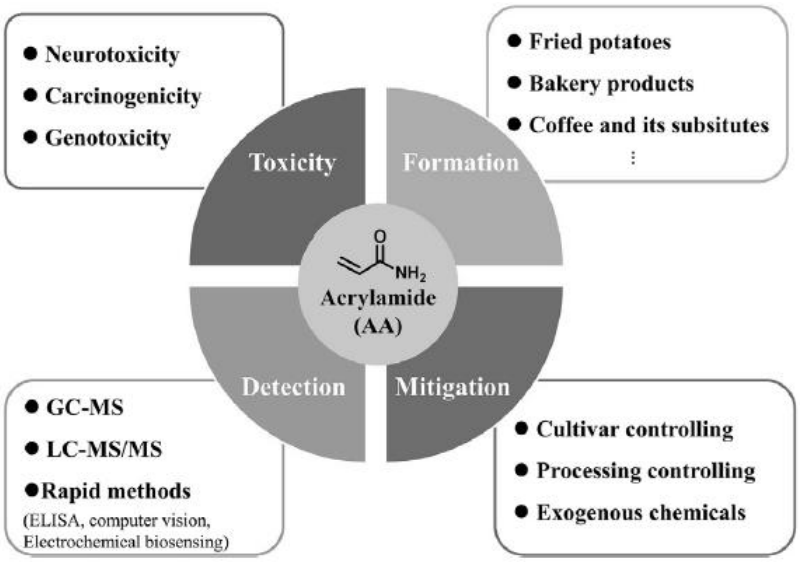


Figure 1: *Recent researches on acrylamide in thermally processed food by Hu et al., (2015)*

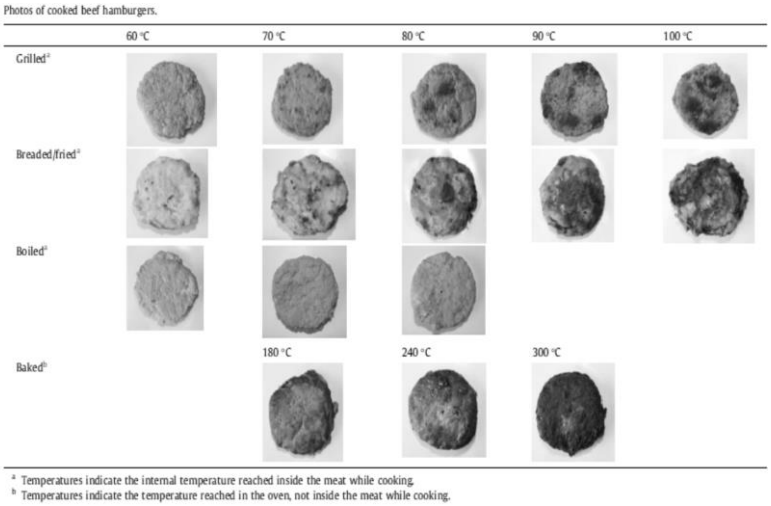


Figure 2. *Trial of Trevisan et al. (2016) for study the effect of temperature changes on acryalmide formation in beef burger*

Table 1. *Measured levels of acrylamide in meat according to Eriksson (2005)*

Type of food (Number of samples/analysis)	**Acrylamide content; median (range)** (µg/kg heated food)
Laboratory-fried protein-rich food	
Beef, minced (5)	17 (15 – 22)
Chicken, minced (2)	28 (16 – 41)
Cod, minced (3)	< 5 (< 5 – 11)

Methods of detection of AA

In April 2002, the formation of acrylamide in starch-rich foods or high-temperature cooking, like with a variety of baked and fried foods cooked at high temperature, was reported by researchers from the Swedish National Food Administration (SNFA) and the University of Stockholm (***Keramat et al., 2010).*** At that time, there was no sufficiently validated method for the detection of acrylamide.

1. Conventional detection methods

Liquid Chromatography-Mass Spectrophotometer (LC-MS/MS-based) method (figure 2) was then developed by various investigators as ***Rosén andHellenäs (2002); Ono et al. (2003); Swiss Federal Office of PublicHealth (2002); Takatsuki et al. (2003); US Food and Drug Administration (2003).***

The main techniques for analyzing AA in food samples are liquid and gas chromatography, both coupled with mass spectrometry (LC- and GC–MS) after extraction (with water or mixtures of water and organic solvents) and clean-up of the food extract (mainly by Solid Phase Extraction (SPE), or by matrix solid-phase dispersion methods (MSPD) or solid phase microextraction (SPME) ***(EFSA, 2015)***.

So far, many analytical methods have been used to quantify acrylamide in food products. Most of them are expensive and demand sophisticated instruments, such as high-performance liquid chromatography (HPLC) ***(Qin et al., 2017).***

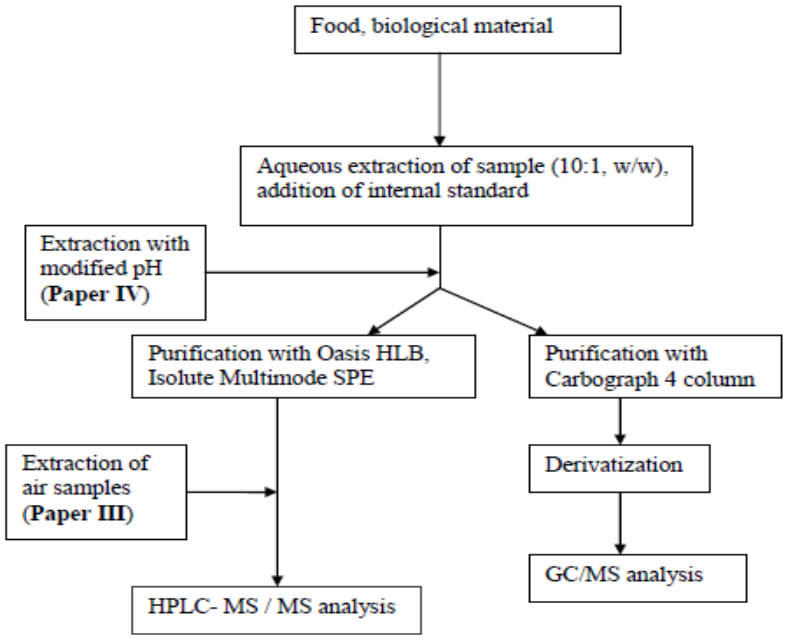


Figure 3. *Summary LC-MS/MS-based method for measurement of acrylamide in food (Eriksson 2005)*

Standard and conventional methods for the detection of AA in Foods Liquid Chromatography (LC) and high performance liquid chromatography (HPLC) coupled with mass spectrometry (MS) are the most preferred methods for separation and quantification of AA in foodsUltra-performance liquid chromatography (UPLC) allows better separation of AA in food matrices within a shorter retention time and higher sensitivity ***(Zhang et al., 2007)***. A UPLC-ESP (electrospray)-MS/MS method was reported for the detection of AA in foods, which supplied a rapid quantitative procedure for AA within a run time of only 3 min, along with good repeatability (RSD _ 4.5%) within a day (n ¼ 5) and day-to-day (n ¼ 10) precision tests ***(Zhang et al., 2007).***

Some papers developed and appliedGas chromatography coupled with electron capture detector (GC–ECD) successfully for the rapid determination of acrylamide in fried foods, such as potato crisps, potato chips, and fried chicken wings ***(Zhang et al., 2006)***. First GC–ECD method dealing with contaminant analysis of acrylamide and the first modified method employing potassium bromate and potassium bromide as derivatization reagents for acrylamide derivatization, the method included defatting with n-hexane, extraction with aqueous solution of sodium

chloride (NaCl), derivatization with potassium bromate (KBrO3) and potassium bromide (KBr), and liquid-liquid extraction with ethyl acetate. The final acrylamide extract was analyzed by GC–ECD for quantification and by GC–MS for confirmation. The chromatographic analysis was performed on the HP-INNOWax capillary column, and good retention and peak response of acrylamide were achieved under the optimal conditions (numbers of theoretical plates N= 83,815).

The limit of detection (LOD) was estimated to be 0.1 _g kg−1 on the basis of the ECD technique. Recoveries of acrylamide from conventional samples spiked at levels of 150, 500 and 1000 _g kg−1 (n = 4 for each level) ranged between 87 and 97% with relative standard deviations (RSD) of less than 4%. Furthermore, the GC–ECD method showed that no clean-up steps of acrylamide derivative would be performed prior to injection and was slightly more sensitive than the MS/MS-based methods. Validation and quantification results demonstrated that this method should be regarded as a new, low-cost, and robust alternative for conventional investigation of acrylamide ***(Zhang et al., 2006)***.

Another simple, fast and cost-effective method was developed by ***Altunay et al. (2016)*** for the indirect determination of acrylamide in processed foods particularly consumed by children. The method is based on ion–pairing of acrylamide with fluorescein (F2_) in presence of Ni(II) ions at pH9.0, and then extraction of the formed ternary complex into a micellar phase of poly(ethyle- neglycol-mono-p-nonylphenylether) (PONPE7.5) before analysis by flame atomic absorption spectrometry (FAAS). In the near past, rapid and reliable liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) method was developed ***Alpozena et al. (2015)*** for the determination of acrylamide in three different local bread types; wheat bread, bran bread, whole wheat bread. Acrylamide analyses were made in crust parts of the 85 bread samples. The method was linear up to 750 μg kg–1 food with a determination coefficient of 0.999. Recovery rate was found 99.3% with limit of detection and limit of quantification values of 1.5 μg kg–1 and 5.0 μg kg–1, respectively. Certified reference materials of crisp bread were analyzed and acrylamide contents of these samples were found in the range cited in the certificates. Statistical correlations were investigated between acrylamide contents and protein contents, reducing sugar contents, moisture contents, pH, and color parameters (L*, a*, b*) of bread samples.

A recent method by ***Lambert et al. (2018)*** based on capillary

electrophoresis (CE) is a powerful alternative for analyzing organic compounds based on charge-to-mass differences with high separation efficiency. Two in-line preconcentration capillary zone electrophoresis (CZE) methods (field amplified sample injection (FASI) and stacking with sample matrix removal (LVSS)) have been evaluated for the analysis of AA in foodstuffs after being derivatized with 2-mercaptobenzoic acid, both of which showed similar sensitivity and precisions compared with chromatography-based methods ***(Bermudo, et al., 2006 and Bermudo et al., 2007)***. A laser-induced fluorescence detection method mediated by CE with CdTe quantum dots (QDs) as amplifier was studied for the detection of AA in potato crisps with good recoveries liquid chromatography coupled with tandem mass spectrometry to quantify acrylamide in foods. The AA content was determined by the reversed phase-high performance liquid chromatography (RP-HPLC) method coupled to a diode array detector (DAD) ***(Michalak et al., 2017)***.

2. Rapid detection methods

Rapid detection methods are mainly based on biochemical properties of AA, biomaterials with high and specific interaction with AA, or changes of physiochemical properties of thermally processed foods related to AA. The standard and rapid methods for detection of AA in foods are summarized in Table 2

The method described by ***Asnaasharia et al. (2018)*** successes in determination of acrylamide even at low concentrations (figure 4). In their study, a simple, rapid and accurate fluorescent sensor was developed for detection of acrylamide based on gold nanoparticles (AuNPs) and FAM-labeled double-stranded DNA (FAM-dsDNA). The sensing method was developed in a way to produce a remarkable fluorescence intensity difference in the absence and presence of acrylamide. In the presence of acrylamide, the single-stranded DNA (ssDNA) and acrylamide adduct are formed. So that, FAM-labeled complementary strand DNA (FAM-csDNA) is free in the environment and adsorbed on the surface of AuNPs and as a result, the FAM is quenched by the AuNPs. Under optimized conditions, the presented fluorescent analytical approach showed high selectivity toward acrylamide with a wide linear response, 1 × 10−7M–0.05 M, and a limit of detection (LOD) of 1 × 10−8M for acrylamide.

Most recently method by ***Yadav et al. (2018)*** improved acrylamide biosensor was developed by immobilizing HbNPs onto paper-based devices. HbNPs based lab on chip acrylamide biosensor showed better analytic performance in terms of lower detection limit (0.1 nM), wider working range (0.1 nMe100

mM). The use of HbNPs not only improved the analytic performance of the biosensor but also simplified its fabrication process, transportable and economic.

Table 2. *Applications of standard methods and rapid methods for detecting AA in thermally processed foods*.

	Method	Sample	Linear range	LOD ($\mu g\ kg^{-1}$)	LOQ ($\mu g\ kg^{-1}$)	Recovery	RSD	Reference
Standard Methods	LC–MS/MS (HPLC, UPLC)	Potato chips	1 - 200	1	3	81.6–99.0%	0.4–4.5%	(Zhang et al., 2007)
		Coffee	2 - 100	5	16	92–95%	<5%	(Bortolomeazzi et al., 2012)
		Cereal-based foods (breakfast cereal, cookies)	1 - 2,000	6	18	90.6–98.5%	1.8% (mean)	(Şenyuva & Gökmen, 2006)
		Tea	1 - 20	1	5	74–79%	1.6–8.3%	(Liu, Zhao, Yuan, Chen, & Hu, 2008)
		Infant foods	0.1-200	1	3	87–96%	<6.5%	(Zhang et al., 2005)
	GC–MS (GC based)	Potato chips	10 -1,000	5	–	81.9–95.7%	5.3–13.4%	(Yamazaki et al., 2012)
		Coffee	0 - 1,500	5	10	84–97%	2–10%	(Soares, Alves, Casal, Oliveira, & Fernandes, 2010)
		Cereal-based foods (biscuits, cracker, breakfast cereals)	5 - 50,000	2	36	91–99%	<4%	(De Vleeschouwer et al., 2007)
		French fries	30-10,000	1	25	>96%	<2%	(Notardonato, Avino, Centola, Cinelli, & Russo, 2013)
		Instant noodles	10 - 5,000	5.1	13	87.3% (mean)	1.6%	(Yamazaki et al., 2012)
Rapid Methods	Electrochemical Biosensors	Potato crisps	9.2×10^{-4} - 3.4×10^{3}	8.5×10^{-3}	–	–	–	(Stobiecka et al., 2007)
		Potato crisps	0.35- 5.3×10^{6}	1.4×10^{-2}	–	95.40–97.56%	–	(Batra, Lata, Sharma et al., 2013)
		No food detected	0.71-710	2.84	7.1	–	–	(Garabagiu & Mihailescu, 2011)
	ELISA	Pringles crisps	51.76-3,311.5	65.7	–	–	–	(Preston et al., 2008)
		Mashed potatoes	50 - 1,280	50	350	92.6–95.5%	–	(Fu et al., 2011)
		Potato crisps, instant noodles, biscuits, and cakes	26.3 - 221.1	18.6	60.6	74.4–98.1%	–	(Quan et al., 2011)
		Potato fries, biscuits	10 - 100,000	6	–	90–110%	6.3–9.9%	(Zhou et al., 2008)
		Potato chips, cookies, and coffee	0.25 - 24.15	0.036	–	73.7–107.7%	3.6–19.2%	(Wu et al., 2014)
	Fluorescence	Potato chips	35 - 350,000	35	–	–	–	(Hu et al., 2014)
		French fries, fried puffs, fried chicken roll, bread, biscuits	50 - 20,000	15	–	66.0–110.6%	–	(Liu et al., 2014)
			Correlation coefficient			**Prediction accuracy**		**Reference**
	Computer vision	Potato chips	0.989			98% at a threshold of 1000 μg/kg		(Gökmen et al., 2006)
		Cookies	0.946			100% at a threshold of 150 μg/kg		(Gökmen et al., 2008)

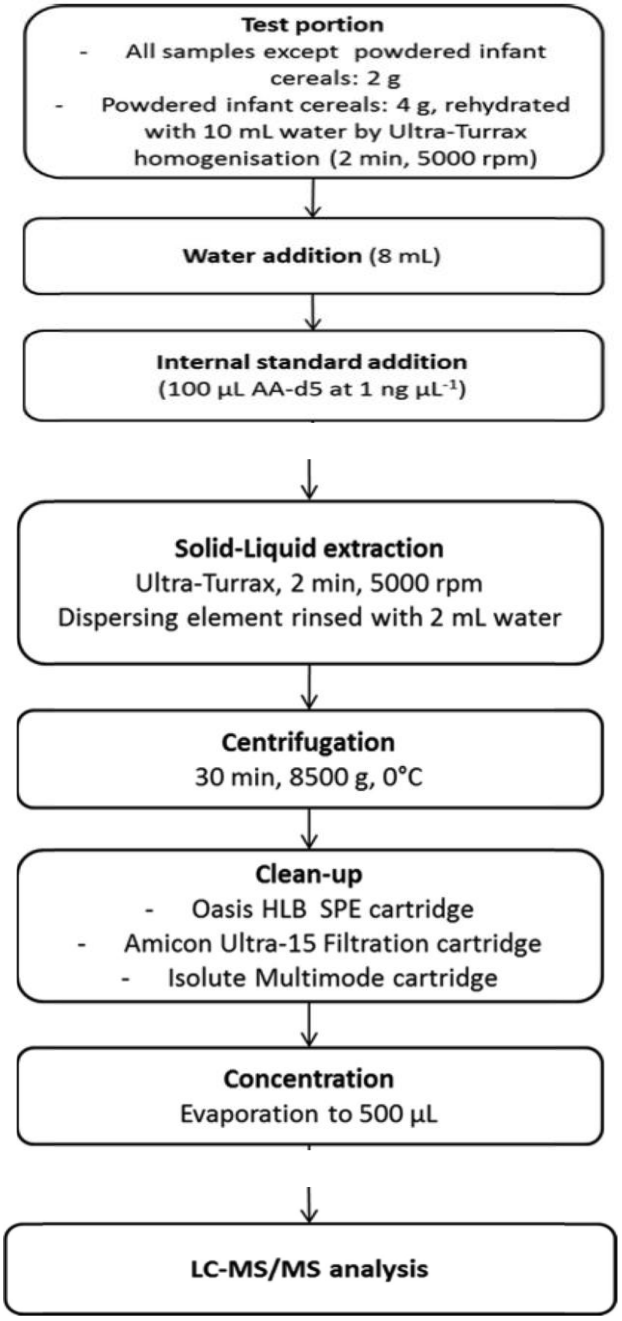
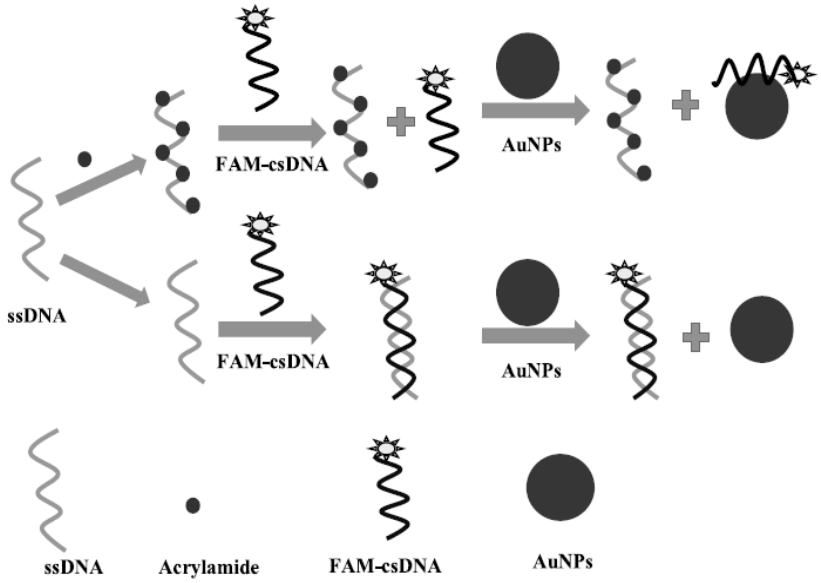


Fig. 4. *Diagram of the sample extraction and purification proceduresaccording to method described by Asnaasharia et al .(2018)*



Scheme 1. Schematic description of acrylamide detection based on the fluorescent biosensor.

By ***Asnaasharia et al. (2018)***

Table 3. *analytical recovery of added acrylamide in processed foods measured by HbNPs/EPAD (Yadav et al., 2018).*

Sr. No.	Acrylamide added (mM)	Acrylamide found (mM)	% Recovery
1	–	45.6	100
2	15	59.8	98.67
3	20	65.0	99.02

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تطور طرق الكشف عن الأكريلاميد في الأطعمة المطهية منذ اكتشافه لأول مرة عام 2002 مقال علمي

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الملخص العربي

إن التغيير في اسلوب الحياة المعاصرة ادي الي ضرورة توفير وجبات جاهزة سريعة تواكب سرعتها مما نتج عنه مخاطر كيميائية بالاغذية نتيجة الي المعالجة الحرارية اثناء الطهي. من ضمن تلك المخاطر الكيمائية مركب الأكريلاميد المتهم في تشوهات الاجنة وتلف الجهاز العصبي يتواجد في مختلف الأطعمة المعالجة حرارياً مثل رقائق البطاطا والبسكويت والخبز. وحتي الحين فأن معظم المراجع العلمية لم تعطي اهتماما لتواجدها في وجبات اللحوم والاسماك المقلية. لذلك يقدم هذا المقال نظرة عامة شاملة للكشف عن الأكريلاميد في الأطعمة المختلفة منذ اكتشافه لأول مرة في أبريل عام 2002 في الوجبات المقلية من قبل باحثين من إدارة الأغذية الوطنية السويدية وجامعة ستوكهولم. وتم استعراض طرق الفحص الحديثة القائمة على تكنولوجيا النانو والبيولوجيا الجزيئية. ويستعرض المقال ايضا طرق معملية عالية الحساسية والانتقائية والاستقرار مثل تقنية الكروماتوغرافيا السائلة إلى جانب تقنية الطيف الكتلي الترادفي. بالاضافة الي طرق كشف سريعة المعتمدة علي برامج الكمبيوتر ومُقَايَسَةُ المُمْتَزّ المَناعِيّ المُرْتَبِطِ بالإنْزيْم والاستشعار البيولوجي الكهروكيميائي والاستشعار البيولوجي الفلورسنت. هذا وقد تم مناقشة كل الطرق والاقتراحات اللازمة لإجراء المزيد من البحوث حول الطرق السريعة للكشف عن مركب الأكريلاميد بناء على أنواع الأطعمة والتحديات التقنية.