Introduction:
Acrylamide is potentially toxic classified as a neurotoxicant, carcinogen and teratogen (IARC, 1994). Acrylamide (AA) is an odorless crystalline solid at room temperature, with molecular formula C3H4N2O and molecular weight of 71.08. The monomeric form of AA is a water-soluble powder and it is employed in different chemical and industrial processes. It is a vinyl monomer and its industrial application is associated with pollution and health risks (Brown et al., 1980). Acrylamide is a component of polymers and co-polymer materials used in gel chromatography and in waste water management (Kopp and Dekant, 2009; Bekas et al., 2006). Acrylamide is also a contaminant in foods prepared at very high temperatures. Acrylamide is formed during high-temperature cooking of animal feed and that is found in common human foods that are prepared by cooking at high temperatures. It has generated interest in the neurotoxicity of dietary exposure to acrylamide. Acrylamide is largely derived from the heat-inducing reactions between the amino group of the amino acid asparagine and carbonyl groups of glucose and fructose in plant-derived foods. The remarkably high levels of acrylamide were observed for the first time by Eriksson (Eriksson, 2005). Direct consumer exposure to acrylamide may result from ingestion of high-carbohydrate foods, such as potato crisps and chips, roasted cereals and breads. In Egypt, there has been an upward trend in consumption of snacks, that as fast foods and drinks, between main meals. (Elhassaneen and Sayed, 2015).

At present, several analytical methods are available for determining acrylamide in foods and the majority are classical methods based on high performance liquid chromatography (HPLC) or gas chromatography (GC) techniques (Singh and Balaji, 2010). But, analysis of acrylamide in food products focuses primarily on chromatographic methods such as HPLC–MS and GC–MS (LC-MS) (Jio, et al., 2005; Hoenicke, et al., 2004). The HPLC method has the advantage that the transfer of acrylamide into an organic solvent or the removal of water after aqueous extraction is not necessary. HPLC has been credited with qualitative and quantitative analysis of various components, biological and non-biological (Matuszewski et al., 2003; Wenzl et al., 2003). Reversed phase (RP)-RP-HPLC-method may be used as a convenient, reliable and low-cost alternative system for liquid and gas chromatography coupled to mass spectrometry methods and can be easily adopted by non-specialized analytical laboratories (Michalak et al., 2013). Unfortunately, there is a dearth of information concerning the quality of these foods and their effects on public health. Therefore, the present study was carried out to determine the concentrations of acrylamide in selected snacks, cereals, bread products and baby food at available markets in Egypt by Photodiode Array (PDA) HPLC.

Materials and Methods:

Chemicals:
Acrylamide (99%) was purchased from Sigma (MO, USA). Acetone, hexan and acetonitrile (min. 99%) for HPLC analytical grade were purchased from El Gomhouria Company for Trading Chemicals and Medical Appliances, Egypt.

Market Samples:
Samples i.e. potato chips, snacks, cereals, baby food and coffee (different brands), which represent the most common brands distributed in the Egyptian local markets, were collected at random from the Qalyoubia governorate, Egypt. All samples were purchased on January-2015, transported to the laboratory and used for acrylamide determination.

Twenty of various foodstuffs divided into seven groups, were analyzed in this study. Group 1: Potato chips 1 and 2 (according to different producers). (a) Potato chips 1 (flavored salt and tomato), (b) Potato chips 2 (flavored pepper and tomato). Crisp potato. Group 2: Maize snacks (flavored cheese and pepper); Group 3: breakfast cereals; Group 4: Bread (a) Arabian white bread, flat, produced from white wheat flour (zero type extraction rate 72%). (b) Arabic dark bread (whole flour is used). (c) Crisp bread (olive and butter); Group 5: Biscuit; Group 6: Coffee (a) Soluble Coffee, (b) Roasted Turkish coffee (Arabica) and Group 7: (a) Powdered baby food, were basically composed of cereal and milk in addition to fruit or honey, (b) Jarred baby food 1 and 2.

Processing and Extraction of Samples:
Samples (potato chips, cereals, crisps) were crushed in a mill cup blender and made homogeneous. Bread and jarred baby food have to be pre-dried carefully using vacuum oven. The sample material was mixed well and then filled into a sample cup.

Extraction was carried out according to the method of Khoshnam et al., 2010. Finely ground and homogenized chips sample (5.0 g) were weighed into a closed flask, defatted twice by adding 10 ml hexane and shaking for 5 min. The mixture was dried under vacuum, after decantation for the extraction of AA. 20 ml of acetone and 100 ml of water were added to the defatted sample. The flask was placed in an ultrasonic bath at 40 °C for about 20 min. The acetone was filtered through a filter paper. Gently, 10 ml of the filtrate was evaporated under vacuum to dryness. Then, 2 ml of water was added and shaken thoroughly to dissolve the residue. The aqueous solution was filtered through a disk filter and subjected to HPLC analysis. For baby food, sample were homogenized and different concentrations of AA standard, was used as spiked samples.

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Acrylamide Content in Selected Commercial Foods in Egypt Using High-Performance Liquid Chromatography–Photodiode Array Detection.
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ABSTRACT
This study reports the results of the survey study on acrylamide levels in selected foods products collected randomly from supermarkets in Egypt. Acrylamide (AA) can be industrially produced or formed in processed food by a Millard type of reaction between sugar molecules and the amino acid asparagine. In order to estimate the quality of these foods and their effects on public health, the content of acrylamide in commercial potato chips, breakfast cereals, snacks, biscuit, bread, coffee and baby food has been determined. High-performance liquid chromatography (HPLC) coupled to photodiode array detector technique was employed for the analysis. Extraction method was followed in order to allow low concentrations of acrylamide to be detected. Limit of detection was 30 μg/kg. In general, the highest value of acrylamide (1226 μg/kg) was detected in potato crisps whereas the lowest value was detected in powdered baby food (20 μg/kg). The outcome of current study has strongly recommended the necessity to conduct a large-scale survey to evaluate the levels of acrylamide in commercial food products.
Identification and Quantitation of Acrylamide:
Using reversed phase (RP) C18-type column (Hypersil ODS C18) of 4.6 mm × 15 cm. Chromatographic analyses were carried out using a Young Lin HPLC, series YL-9100, equipped with a quaternary pump, an autosampler (YL9150), a degasser, and a YL-9160 spectrophotometric detector (Photo Diode Array detector – PDA), which was set at 202 nm. The flow rate was fixed to 0.5 ml/min. The solvent system consisted of acetonitrile /H2O (70:30). Mode of the HPLC instrument was isocratic and injection volume was 20 μl.

Results and Discussions:
Acrylamide determined in selected commercial samples in 2015. Different methods were reported for the extraction of acrylamide. These could be divided into two main categories extraction with water or with an organic solvent (Khoshnama et al., 2010). With respect to the applied HPLC method, in all cases the extracted AA was finally transferred to an aqueous phase prior to injection. Acetone was used to extract the AA from samples. Acetone yield much more clear extract than that of water. In addition, it can be easily evaporated under a gentle stream of nitrogen. In addition, using acetone for extraction gave minimum interference (Khoshnama et al., 2010; Takatsuki et al., 2003).

Processed potato and cereal based food samples usually composed of high amount of colloids and fat, which should be have de-fatting step before extraction step (Pettersson et al., 2006). The use of defatting with n-hexane had in most cases no significant effect on the acrylamide yield, not even for fatty matrices (Pengyan et al., 2008) Defatting was accomplished in the same tube as the extraction.

Calibration Curve:
Standard acrylamide was determined with peak observed at 3.5. A good coefficient of correlation of 0.9977 was estimated (Figure 1). The limit of LOD (signal to noise 3) 10.5 μg/kg was detected, while the limit of quantification LOQ (signal to noise 10) was 33.3 μg/kg. Laboratory experiments showed that its recovery was at level of 85% ±5%.

The analysis of potato chips was done, 14 characteristic peaks were observed. Figure 2 show chromatogram of potato chips spiked with acrylamide. Two corn snacks products were analyzed 11characteristic peaks observed. Figure 3 a, show corn flakes chromatogram. The same sample spiked with acrylamide at level 2000 μg/kg (Figure 3 b) . Acrylamide was eluted at 3.5 min.

<table>
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<th>Table 1: Acrylamide level in selected foods and drinks in Egypt.</th>
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The levels of AA varied considerably between single foodstuffs within food groups (Table 1). AA levels in food and drink groups (according to the highest level in comparative samples within the same group). The potato chips group contained the highest level of acrylamide compared with the other six groups. In potato chips group, highest level of acrylamide has been found in crisp potato (1226 μg/kg ) followed by potato chips 2-flavored tomato (773 μg/kg). The lowest level of acrylamide in potato chips group was found in potato chips 1-flavored salt (560 μg/kg). Level of AA in corn snacks group ranged from 553 μg/kg in corn flavored cheese to 620 μg/kg in corn flavored cheese pepper (Table 1). In the breakfast cereals group, corn flakes had moderate level of acrylamide 354 μg/kg and 233μg/kg. Bread group had a variation in AA levels, crisp bread butter was the highest level (528 μg/kg) while Arabian white bread was the lowest level (70 μg/kg). In addition, biscuit had 170 μg/kg acrylamide. Moreover, AA level in soluble coffee was 420 μg/kg while roasted Turkish coffee had lower amount of AA (130 μg/kg). In baby food group AA level ranged from 20 μg/kg for cereal baby foods to 56 μg/kg for jarred baby foods).

The levels of AA in the data presented in Egyptian markets are comparable with previous published data in other countries (Table 1). Highest level of acrylamide has been found in potato crisp. The increasing amount of AA may be due to processing implemented by industry. Potato chips and snacks had lower amount of acrylamide. The present results are in agreement with previous survey studies on AA level (Svensson et al., 2003; Bekas et al., 2006 and Moiska et al., 2012). Formation of acrylamide in potato chip fried traditionally or under vacuum increase either with time or temperature (Granda and Moreira, 2005). Cereals have differing potential for the formation of AA, depending on their type and varying content of free asparagine. Raw materials with low asparagine contents cause the extrusion process to form end products with low AA values.

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Food group | Food subgroup | AA level µg / Kg | AA level µg / Kg in previous published data
--- | --- | --- | ---
Potato chips | Chips1 flavored salt | 560 | 468-656 (U.S. Food and Drug Administration 2003; 380-755 (Bekas et al., 2006); 14-40 (Brill et al., 2005 ))
| Chips1 flavored tomato | 646 | 605 | 560-656 (U.S. Food and Drug Administration 2003; 380-755 (Bekas et al., 2006); 14-40 (Brill et al., 2005 ))
| Chips 2 flavored pepper | 773 | 693 (U.S. Food and Drug Administration, 2003)
| Chips 2 flavored tomato | 773 | 693 (U.S. Food and Drug Administration, 2003)
| Crisp potato | 1226 | NI:
| Corn snacks | Corn flavored pepper | 620 | NI |
| Corn flavored cheese | 553 | NI |
| Breakfast cereals | Corn flakes1 | 233 | 77 (U.S. Food and Drug Administration, 2003) |
| Corn flakes2 | 354 | 77 (U.S. Food and Drug Administration, 2003) |
| Bread | Arabian white bread | 70 | 180 (Al-Dmoor et al., 2004); 90 (El-Ziney et al., 2009)) |
| Arabian dark bread | 58 | 40 (El-Ziney et al., 2009) |
| Crisp bread butter | 528 | 427 (Valentina et al., 2012) |
| Crisp bread olive | 196 | |
| Biscuits | Biscuit fingers | 170 | 75-2520 (EFSA, 2010) |
| Coffee | Soluble Coffee | 420 | 240-359 (U.S. Food and Drug Administration, 2003) |
| Roasted Turkish Coffee | 130 | 351 (Friedman, 2003); 259 (Svensson et al., 2003) |
| Powdered baby food cereal and milk | Cereal baby food1 | 45 | 6.8-124.9 (Jio et al., 2005); 69-353 (EFSA 2010) |
| Cereal baby food2 | 20 | |
| Jarred baby food1 | 56 | 44-162 (EFSA, 2010) |
| Jarred baby food2 | 40 | |

NI: No information

Acrylamide is formed in coffee beans due to Maillard reaction during roasting that takes place at high temperature, for example, in range of 220-25°C. The degree and speed of roasting affects acrylamide formation due to formation kinetics (Lantz et al., 2006). Roasting causes acrylamide to form in coffee beans beyond the commonly accepted asparagine/sugar condensation (Guenther et al., 2007).

Crisp bread had the highest level of acrylamide in bread group. While white bread contained higher AA than dark bread. However, AA formation is varied depending on flour type, ingredients, temperature time and layer thickness. Lowest concentration of AA was observed in baby food in current study. Jio et al. (2005) reported AA level in cereal baby food from 6.8 to 124.9. EFSA (2010) reported that the arithmetic mean acrylamide content ranged from 44 to 353 µg/Kg for 'jarred baby foods. This variation in current study and also the variation in other reported studies related to acrylamide formation factors such as content of reducing sugar, free asparagine, industrial processing including temperature, pH and surface to volume ratio of food material.

However, FAO and WHO (2002) recommended that, AA presumably would be present in acceptable levels in nutritionally balanced diet levels in individual foods should be as low as reason-ably achievable.

**Conclusion:**
Acrylamide is formed via the Maillard reaction between reducing sugars and asparagine in carbohydrate-rich foods during heat treatment processes such as frying, baking, roasting and extrusion. This study used HPLC methodology for the quantitative analysis of acrylamide in baby food cereals and baby food in jars. This method is successfully applied to the quantification of acrylamide in baby foods, which had low acrylamide content 20-56 μg/kg.

A survey of selected commercial foods in Egypt, foods from the Egypt market was performed for determining the AA contents in these products. The highest level of AA was determined in Potato crisps and chips. In general, snacks, cereals, crisp bread and soluble coffee were among the food products with high levels of AA. Significant differences were observed in the AA contents of different brands of the baby food, chips and cereal. Moreover, it is highly required to establish AA determination method. RP-HPLC-PDA method used as a convenient, reliable and low-cost method and can be easily adopted by non-specialized analytical laboratories. Therefore, the present study monitored the concentrations of acrylamide in selected products and baby foods at available markets in Egypt by HPLC.
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