

Determination of Monosodium Glutamate Content in Selected Traditional Meat Dishes

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Abstract:

The study was conducted to determine the monosodium glutamate (MSG, Aginomotto or Chinese salt) content of selected traditional meat dishes. Six traditional meat dishes were selected from five different restaurant of Lahore. The dishes included were chicken karahi, mutton qorma, chicken biryani, seekh kabab, chicken tikka, palak gosht from five different places, which were Mazang, Lakshami Square, Fortress Stadium, Johar Town, M.M Alam Road. Estimation of monosodium glutamate was carried out by modified HPLC (High Performance Liquid Chromatography) method of Lateef, Siddique, Saleem, & Iqbal, 2012 with minor modification. HPLC procedure included solution preparation, PH determination, Derivatization, Acidification, Extraction, Sample injection. The peak of MSG was identified by comparing it with retention time of MSG standards, that is, 8.2 min. The results from laboratory analysis concluded that MSG was present, altogether in the traditional meat dishes excluding five which were palak gosht from Lakshami Square, chicken tikka, chicken karahi, seekh kabab from Johar Town and mutton qorma from M.M Alam Road. On the bases of the data it was concluded that each restaurant adds MSG to makes their food more flavorful every time, it was observed that all the representative samples of chicken tikka and chicken biryani contains MSG. It was also concluded that the amount of MSG was with the optimal palatability concentration i.e. 0.2 – 0.8% (w/w) as suggested by Loliger in 2000.

Key words: MSG, HPLC, Traditional Meat Dishes, Retention Time, Peak of MSG, Derivatization, Tolerable Upper limit

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1. INTRODUCTION:

Monosodium glutamate (MSG, Aginomotto or Chinese salt) is a food additive and has E621 code commonly marked as flavor enhancer (European Food Information Council, 2006). Around the world monosodium glutamate is used habitually, it adds flavor to the canned chicken broth, packs of onion, sauces, soup mix, cheese and the low-fat yogurts (Moskin, 2008).

It does not enhance the four basic tastes (bitter, salty, sour, sweet) but it does enhance the complex flavors of meat, poultry, seafood, and vegetables by elevating the taste buds with a flavor known as UMAMI, which is experienced as a meat or broth like taste (Fact on monosodium glutamate, 2002).

Monosodium glutamate is an excitotoxin, which basically means, a chemical substance that excites your neurons and may cause death. It may aggravate many neurological disorders such as Alzheimer's and Parkinson's disease. MSG side effects may include seizures, brain cell damage, allergies, rashes, asthma attacks, headaches, and brain tumors (Blaylock, 1996). In some recently conducted studies, the most frequently reported symptoms were headache, numbness/tingling, flushing, muscle tightness and generalized weakness. Prevalence of these symptoms are suggested to be 1– 2 % of the general population (Yang, Drouin & Herbert, 1997; Geha et al., 2000).

The monosodium glutamate dosage that produced reactions in human being ranged from 0.5 grams to 2.5 grams (FDA, 1995). The optimal palatability concentration for MSG is between 0.2 – 0.8% (w/w) and its use tends to be self-limiting as over-use decreases its palatability (Loliger, 2000). The largest palatable dose for humans is about 60 mg/kg body weight (Walker & Lupien, 2000).

However, there are some studies showing certain amount of MSG has possible toxic effects and may be associated with myocardial and hepatic diseases (Ault, 2004). Therefore, scientific literature describing 'MSG symptom complex'. Regarding this, a Federation of

American Societies for Experimental Biology (FASEB) compiled a report on behalf of FDA that MSG as a food ingredient has been the subject of health studies. In the late 1960s, numerous case reports appeared in the sci that concludes that MSG was safe for most people when eaten at customary levels. However, it also said that, based on anecdotal reports, some people may have MSG intolerance. 'MSG symptom complex' which are considered representative of the acute, temporary and self-limited reactions to oral ingestion of MSG (FASEB, 1995).

The Chinese have used it for centuries to deepen and bring out the natural flavors of foods, but now due to its hazardous effects which have become prominent, they start avoiding it. It can enhance the taste, especially protein rich items. If top quality, fresh ingredients are used, MSG is not necessary (Giacometti, 1979). Addition of monosodium glutamate increases day by day in traditional continental meat dishes served over diverse restaurants, and would have more destructive effect for the reason that these dishes does not have any constituent list or a labeled table, which would direct the consumer (Reejhsinghani, 2005). It is predicted that its injurious effects also may seen in Pakistan within few years only because of its excessive usage in a variety of traditional cuisine.

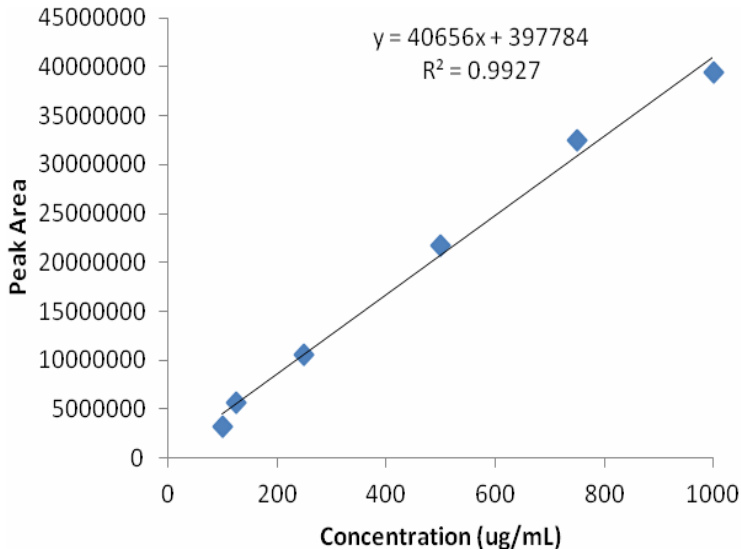
2. Research Methodology

2.1 Apparatus:

Conical Flask, Decanting (Separating) Funnel, Volumetric Flask, Beaker, Pipette, Stirrer, Thermometer, HPLC (High Performance Liquid Chromatography), Perkin Elmer, 200 Series, Digital Weighing balance, UV Spectrophotometer, Sonicator

2.2 Reagents

MSG standard (99%), 2, 4, dinitro-1-fluorobenzene (DNFB) (a kind gift from Dr. Kausar Siddiq PCSIR Labs, Karachi), Sodium bicarbonate (5 % w/v), Hydrochloric acid (6M), diethyl ether and methanol. All the reagents used were of analytical grade except HPLC grade methanol.



2.3 Preparation of Reagents or Chemicals

Hydrochloric acid (HCL) (6M): 1.08mg of HCL / 5ml of distilled water to form 6M solution of Hydrochloric acid, Sodium bicarbonate solution: 5grams of sodium bicarbonate was diluted in 5ml of distilled water in flask

2.4 Raw Material / Sample description

Six traditional meat dishes were selected as a sample for analysis from five different restaurants which poles apart in Lahore Products included were:

Chicken Karahi, Mutton Qorma, Chicken Biryani, SeekhKabbab, Chicken Tikkah, PalakGosht

Products included were taken from the restaurants of following places of Lahore:

M.M Alam Road, Lahore, Fortress Stadium, Lahore, Johar Town, Lahore, Lakshami Square, Lahore, Mazang, Lahore

2.5 Optimization of Solvent System / Optimum HPLC solvent

Rodriguez used acetonitrile: glacial acetic acid 1% (v/v, 1: 3) as the mobile phase. LATEEF *et al* in 2012 selected, 50% methanol: 50% water, which was compatible with their system. After trying many reported mobile phases (data not given), with minor amendment the second system was the most efficient mobile phase to quantify the monosodium glutamate. Therefore we select 70% methanol: 30% water as compatible with our system for chromatographic separation of MSG in all the samples.

2.6 Preparation of Standard Curve

Stock standard of MSG (5 mg/mL) was prepared in deionized water. The stock was diluted to make the working standard solution which was further diluted serially in deionized water to obtain 1.0, 0.75, 0.5, 0.25, 0.125 and 0.1 mg / mL (1000, 750, 500, 250, 125 and 100 µg/mL) of MSG to make standard curve. pH was adjusted to 7.8 using sodium bicarbonate (5 % w/v). Took 2ml of each prepared concentration solution, 10ul of DNFB was added to derivatize the protein content of the monosodium glutamate Standards were kept in dark for 3 hours. Extraction was carried out with diethyl ether to remove the excess DNFB. 6M of HCl was used to acidify the remaining aqueous solution, as traces of ether evaporate.

The remaining residue was collected with 500ul methanol and 20ul of each standard was subjected to HPLC (High Performance Liquid Chromatography) for chromatographic analysis.

All the running conditions were kept same for all samples and standards. The retention time of MSG standard as well as MSG samples was 8.2 min in certain optimized conditions.

2.7 Standard curve

The standard curve was plotted between peak areas versus different concentrations of MSG standard in µg /mL as shown in Fig.

The regression equation calculated was ($y = 40656x - 39778$) with the correlation coefficient $r^2 = 0.992$.

2.8 Calculation

The absorbance was determined by the spectrophotometer. The concentration was calculated as µg/L, but unfortunately these values were not up to the mark so later standards were injected to HPLC (High Performance Liquid Chromatography) to get an accurate and desirable results.

3. Pretesting

For the purpose of pretesting a known amount (0.5mg) of monosodium glutamate was added in a homemade kabab to verify either this specific amino acid is heat sensitive or not, the kabab was shallow fried and 5mg/ml solution in deionized water was prepared, the 10ml solution were collected and the pH was adjusted to 7.8 afterwards same procedure of standard preparation was adapted to derivatize the amino acid content.

3.1 Quantification of MSG in samples for Analysis

In the laboratory, the amount of MSG (Ppm) was analyzed in meat dishes samples according to procedure given by Pakistan. J. Chem. Soc. Pak., Vol. 34, No.1, 2012

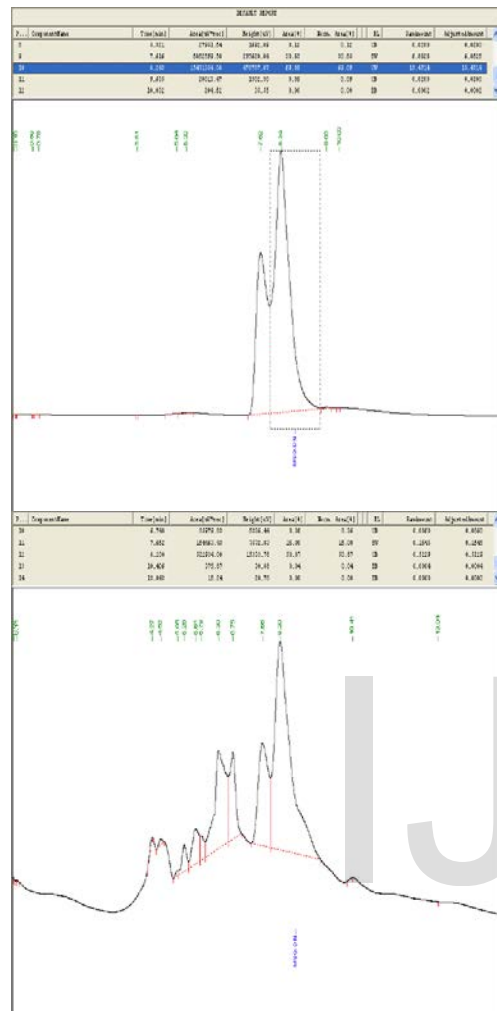
Thirty sample dishes were weighed to make 5 mg / mL solution in deionized water and the 10 mL solutions were filtered by Whatman filter paper. The filtrates containing isolated MSG were collected, to adjust the sample's solution pH range to 7.8 The electrode of the PH meter was washed and dipped into deionized water Then electrode was dipped in the solution reference. pH meter reading was observed when constant. Sodium bicarbonate (5% w/v) was used to change the PH range from acidic to basic.

Both the standard MSG and the MSG isolated from samples needed pre-column derivatization. DNFB (dinitrofluorobenzene) was used as derivatizing agent. Aliquot of 0.5 ml of standard solutions and samples were transferred to a test tube. 10 µL of DNFB was added. The mixtures were placed in a dark box for 3 hours at 30°C. Aqueous solution was acidified with 50 µL hydrochloric acid (6M). Diethyl ether was chosen for extracting excess dinitrofluorobenzene (DNFB), The extraction was performed by taking 0.5-1 ml of diethyl ether for each sample, extraction continued 3-4 times or until the ether no longer gives color. The traces of ether were evaporated until 1 ml was left before analysis. The leftover residue was collected with 500 µL methanol out of which 20 µL of each sample and standard was injected for chromatographic analysis.

3.2 Analysis on HPLC

Samples were run on HPLC Shimpac CLC-OD reversed phase C18 analytical column (83 mm x 4.6 mm i.d, 3 µm) with automatic injector and a 20 µL loop and a UV/ visible detector was used. Sonication of samples and standards was performed by sonicator while the pH meter (PCSIR, Pakistan). Samples were separated with Mobile Phase, consisting of Methanol:Water (70:30) with a Flow Rate 0.5 ml/min and the peak was Detected at UV at 254nm

The samples were used for analysis by HPLC. The samples were then injected into the instrument. Peaks for the monosodium glutamate (MSG) were identified by retention time. Peak areas were used for quantitative analysis.



4. Calculation

Monosodium glutamate was calculated with the help of the chromatograms. The formula is as follows:

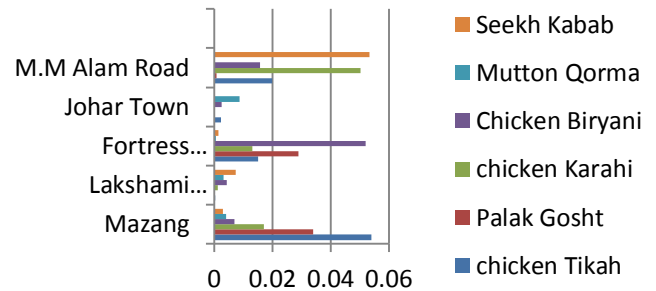
$$\frac{\text{Sample Peak Area}}{\text{Standard Peak Area}} \times \text{Concentration of the Standard} \times \frac{1}{\text{Dilution Factor}}$$

5. Conclusion:

In the laboratory, the amount of MSG (Ppm) was analyzed in all the selected samples of meat dishes according to procedure given by Pakistan. J. Chem. Soc. Pak., Vol. 34, No.1, 2012. The results from laboratory analysis concluded following findings.

MSG was detected in all the meat dishes except five which were palakgosht from lakshami square, chicken Tikkha, chicken karahi, seekh kabab from the restaurant of Johar Town and mutton qorma from M.M Alam Road. On the basis of the data it has been proven that every restaurant adds this flavor enhancing agent in their different cuisines which makes their customers to enjoy the same exotic flavor again and again, especially for chicken tikkah and chicken biryani MSG acts as a most important ingredient as it was detected in the representative samples of both cooked products. It was also found

that the levels of MSG were within the optimal palatability concentration for MSG i.e. 0.2 – 0.8% (w/w) as suggested by Loliger in 2000, therefore they may not lead to any health complications such as “MSG symptom complex” and they are safe to consume in appropriate quantity at proper time. We used a simple and rapid technique to separate MSG from the traditional meat dishes which is reproducible and accessible to be used in quality control food laboratory. The findings were analyzed statistically by using percentage and average and were presented in the form of tables and graphs. Microsoft Word and Excel were used to tabulate the results.



Acknowledgements

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