NUKLEONIKA 2009;54(2):99–105

ORIGINAL PAPER

Detection of radiation treatment of dry plant extracts by thermoluminescence and pulsed photostimulated luminescence. Comparative study*

Katarzyna Lehner, Kazimiera Malec-Czechowska, Wacław Stachowicz, Grzegorz Guzik

Abstract. Results of the examination of the variety of dry plant extracts (Thyme extract, Celery seed extract, Artichoke extract, Citrus aurantium extract and others) by two different detection methods are described. Both PSL and TL methods are presented and discussed. Comparative study based on the analysis of the results obtained by thermoluminescence (TL) and photostimulated luminescence (PSL) measurements delivered the arguments that preselection of detection methods based on model studies is rational to be adapted in analytical laboratories specialized in the detection of irradiated foods.

Key words: plant extract • detection • irradiation • thermoluminescence • photostimulated luminescence

Introduction

Plant extracts are today widely used in the food industry (modification of sensoring features of foodstuffs, diet supplements) as well as in the cosmetic industry (new generation cosmetics).

However, similarly to most of the foodstuffs, fresh and dry products containing plant extracts are typically stored at moderate temperatures to save their unique properties and for that reason may contain living moulds, pathogenic microorganisms as well as eggs of insects and larvae. Dry plant extracts themselves can be contaminated with pathogens, too.

In order to avoid contamination and spoilage of dry plant extracts during their storage, ionizing radiation, an effective tool capable to kill pathogens, is used parallel to other preservation methods. The international trade of irradiated food is not fully controlled and depends on local decision of each country. The European Parliament and the Council adopted two Directives no. 1999/2/EC and no. 1999/3/EC to harmonize the rules concerning the treatment and trade of irradiated foods in EU countries [2, 3]. In view of the above regulation the list of irradiated food products accepted currently for free distribution in the EU market comprises dried aromatic herbs, spices and vegetable seasonings only.

K. Lehner✉, K. Malec-Czechowska, W. Stachowicz, G. Guzik
Laboratory for Detection of Irradiated Food,
Institute of Nuclear Chemistry and Technology,
16 Dorodna Str., 03-195 Warsaw, Poland,
Tel.: +48 22 5041066, Fax: +48 22 5041237,
E-mail: slinz@orange.ichtj.waw.pl

Received: 27 October 2008
Accepted: 18 February 2009

* Presented on the International Conference on Recent Developments and Applications of Nuclear Technologies, 15-17 September 2008, Białowieża, Poland.

It is obligatorily required that irradiated food has to be labelled in addition.

As to our knowledge, there are not many literature data available on the detection of irradiation in dry plant extracts. Certain plant extracts together with other foodstuffs have been only examined by TL and PSL in our earlier comparative study [4]. Plan extracts are usually examined whether irradiated by employing the TL method. The main analytical problem with powdered extracts lies with effective isolation of suitable volume of silicate minerals from investigated product, indispensable to proceed successfully further detection of irradiation and final classification of samples. Having enough of silicate mineral isolated, one will be able to identify radiation treatment by the TL method with a truly high reliability. The aim of the present comparative study is to test whether the PSL method, much simpler and faster than the TL method, could be alternatively used for the detection of irradiation in plant extracts, too. It has to be noted that the PSL method is quite successfully used for the detection of irradiation in herbs and spices.

The results of PSL examination of selected samples of dry plant extracts (Thyme extract, Celery seed extract, Artichoke extract, Citrus aurantium extract and others) compared with TL data are presented and discussed below.

Materials and methods

Preparation of samples for TL analysis

At least 50 g of a sample was suspended in about 500 ml of demineralized water or, if necessary, in another solvent, e.g. methanol. The analytical procedure of the isolation of silicate minerals from dry extracts of herbs, spices, vegetables and fruits selected for the present study was based on EN 1788:2002 European Standard (Polish authorized translation) [6].

TL measurements

Thermoluminescence of a mineral fraction was measured with the use of a computer operated TL reader, type TL/OSL, model TL-DA-15, Risø National Laboratory, Denmark, under the following operational conditions: initial temperature 50°C, final temperature 500°C, heating rate 6°C/s.

The glow curves (Glow 1) of the mineral fraction isolated from the samples were recorded and then, for the purpose of normalization, the mineral fraction fixed in steel cups was irradiated with 1 kGy of gamma rays in a ^{60}Co source "Issledovatel". Then, on the next day, the glow curves were recorded for the second time (Glow 2) under the same measuring conditions as in the first run. For each sample two duplicate measurements were done.

PSL measurements

The PSL measurements of the tested samples that did not undergo any analytical treatments before, have been achieved with the use of a computer-operated PSL analyzer SURRC PPSL Irradiated Food Screening System, Glasgow, Scotland.

Samples of dry plant extracts weighing 2–5 g were dispensed in clean Petri-dishes with a volume suitable to cover completely the lower surface of each. Petri-dishes with samples inside were covered by storage with lids to avoid the contamination of minerals from air. After check in of the PSL apparatus by running an empty chamber test to prove whether it is clean enough, the test with irradiated and non-irradiated standard (paprika powder supplied by SURRC) was accomplished. Subsequently, Petri-dish with plant extract samples inside were introduced into the chamber of the SURRC PPSL system and measured.

The methodology of PSL measurement comprises screening measurements to establish roughly the status of the sample and the second, the so-called calibrated measurement which is conducted after the sample will be exposed to the normalizing dose of 1 kGy of gamma rays. Such a procedure allows to evaluate the PSL sensitivity of the sample and to obtain the final, more reliable result of examination. The criterion of the identification of radiation treatment in PSL is based on two threshold values, the lower $T_1 = 700$ counts/60 s and the upper $T_2 = 5000$ counts/60 s. PSL intensity below the lower threshold indicates that the sample is presumably non-irradiated while PSL intensity exceeding markedly the upper threshold value is regarded as derived from irradiated samples. The intensity that lies between two thresholds and defined intermediate, cannot be used for identification of irradiation. Further examination of the sample has to be done with the use of more reliable TL method. Samples identified as irradiated should be characterized by a negligible or small increase of PSL intensity after normalizing radiation exposure, whereas not irradiated samples (low intensity recorded by screening examination) prove a relatively great increase of the PSL intensity after normalizing irradiation. The PSL measurements are based on the procedures given in the PN-EN 13751:2007 standard (authorized Polish version of EN 13751) [7].

Results and discussion

Table 1 compiles the list of 16 plant extracts examined at the first stage of the study with samples classified earlier by the TL method as irradiated. The results of the PSL examination by both screening and calibrated runs are listed below.

Screening PPSL measurements on 16 samples resulted in the classification of 7 samples as treated with ionizing radiation. These were: Lemon balm extract, Bee balm extract, Olive extract, Artichoke extract, Mulberry extract, Celery seed extract and Mulberry powder. Luminescence intensity of Lemon balm extract was 5354 counts/60 s and 6245 counts/60 s, while that from Bee balm extract was equal to 5066 counts/60 s and 6115 counts/60 s, respectively. Count numbers in both cases are slightly higher than the upper threshold value $T_2 =$ 5000 counts/60 s for both pairs of samples. Similarly, the Olive extract sample shows the intensities of 7187 counts/60 s and 8110 counts/60 s, again higher than T_2. One sample of the pair Artichoke extract sample shows

Table 1. Photostimulated luminescence measurements of dry plant extracts examined by the TL method and identified as irradiated

Number of sample	Name of the product	Screening PSL	Counts/60 s	Calibrated PSL*	Counts/60 s	Identification of the sample by the PSL method
1	Mulberry extract	positive positive	13,094 20,557	positive positive	23,348 22,477	irradiated
2	Lemon balm extract	positive positive	5354 6245	positive positive	8827 6339	irradiated
3	Bee balm extract	positive positive	5066 6115	positive positive	13,204 9704	irradiated
4	Celery seed extract	positive positive	66,780 73,834	positive positive	94,345 126,543	irradiated
5	Artichoke extract	positive positive	6199 13,292	positive positive	11,237 17,321	irradiated
6	Asparagus extract	intermediate intermediate	2943 2338	positive positive	12,108 9066	not classified
7	Marigold extract**	intermediate intermediate	1644 1314	positive positive	10,091 15,994	not classified
8	Olive extract	positive positive	7187 8110	positive positive	8592 12,139	irradiated
9	Thyme extract**	intermediate intermediate	2346 4128	positive positive	5489 5658	not classified
10	Marigold extract**	intermediate intermediate	987 2871	positive positive	14,779 9837	not classified
11	Olive leaf extract	intermediate intermediate	1204 2786	positive positive	14,124 7393	not classified
12	Thyme extract**	intermediate intermediate	1585 2367	positive positive	14,475 6567	not classified
13	Thyme extract**	intermediate intermediate	1469 1290	positive positive	12,903 5390	not classified
14	Melilot extract	negative negative negative negative	399 506 428 428	intermediate intermediate intermediate positive	4629 4511 4906 5181	not classified
15	Mulberry powder	positive positive	131,520 104,874	positive positive	195,010 180,674	irradiated
16	Silibina	negative negative	404 291	intermediate intermediate	1343 1549	not classified

* – after applying 1 kGy normalizing irradiation.
** – samples of the same names, but obtained from different sources.

the intensity slightly higher than T_2 (6199 counts/60 s) while the second one the intensity markedly higher (13,292 counts/60 s). The samples of Mulberry extract and Celery seed extract were characterized by a higher intensity of luminescence (13,094 counts/60 s; 20,557 counts/60 s) and (66,780 counts/60 s; 73,834 counts/60 s), respectively. The highest intensity was observed with Mulberry powder (131,520 counts/60 s and 104,874 counts/60 s) being by two orders of magnitude higher than those obtained with other products examined through this study.

Intermediate results were obtained with another 7 samples out of the 16 samples examined in total and are characterized below.

Asparagus extract (2943 counts/60 s; 2338 counts/60 s), Thyme extract, Marigold extract (1644 counts/60 s; 1314 counts/60 s), Olive leaf extract (1204 counts/60 s; 2786 counts/60 s). With the sample of Thyme extract the examination was repeated three times (no. 9, 12, 13 in Table 1). All the three samples exhibit the luminescence intensities between 700 counts/60 s and 5000 counts/60 s and are classified as intermediate.

The negative result was obtained with two samples. These were: Melilot extract (no. 14 in Table 1) and Silibina (no. 16 in Table 1). In both cases luminescence intensity was lower than 500 counts/60 s i.e. 399 counts/60 s – 506 counts/60 s for Melilot extract and 404 counts/60 s – 291 counts/60 s for Silibina. The ex-

amination of Melilot extract was repeated 4 times (see Table 1).

The next step of PSL examination was a normalizing irradiation of the samples with a dose of 1 kGy in a ^{60}Co source "Issledovatel" in order to follow the calibrated PSL measurements.

Positive results after normalizing irradiation have been obtained with 14 samples, while intermediate results with another two samples of Melilot extract and Silibina.

The final classification of samples by the PSL method is based on the results of both screening and calibration runs. The sample is classified as irradiated, if the result of screening examination is positive, while the luminescence intensity of calibrated examination is slightly higher than that obtained by screening examination, i.e. is of the same order of magnitude or is by one order of magnitude higher [7]. The results that fulfilled the above requirement were obtained with 7 samples. These were: Mulberry extract, Lemon balm extract, Bee balm extract, Celery seed extract, Artichoke extract, Olive extract, Mulberry powder. Therefore, the samples were classified as irradiated.

In case of samples of Asparagus extract, Thyme extract, Marigold extract, Olive leaf extract the evaluation was not so clear. This is because the above samples delivered intermediate results in a screening run, while positive results after calibrated examination. The difference between the luminescence intensities of calibrated and screening examinations is not very high. For example, in the case of Thyme extract this difference is relatively low (no. 9 in Table 1) – screening run 2346 counts/60 s and 4128 counts/60 s; calibrated run 5489 counts/60 s and 5658 counts/60 s). In the case of the second Thyme extract tested (no. 12 in Table 1) the numbers are more differentiated since the screening run 1585 counts/60 s and 2367 counts/60 s, calibrated run 14,475 counts/60 s and 6567 counts/60 s. The third Thyme extract (no. 13 in Table 1) examined delivered for screening run 1469 counts/60 s and 1290 counts/60 s, while for the calibrated one 12,903 counts/60 s and 5390 counts/60 s and the difference is markedly higher. The most pronounced difference is observed with the sample of Marigold extract (no. 7 and 10 in Table 1). The numbers are as follows: first Marigold extract tested (no. 10 in Table 1) screening run 987 counts/60 s and 2871 counts/60 s while calibrated run 14,779 counts/60 s and 9837 counts/60 s. The cases with calibrated luminescence intensities exceeding markedly, by one or two orders of magnitude, the results of screening run could be interpret as resultant from the analysis of a sample that is a mixture of both irradiated and not irradiated product. Nevertheless, reliable classification of these kind of samples is not possible indeed.

Samples of Melilot extract (no. 14 in Table 1) and Silibina (no. 16 in Table 1) delivered negative results in the screening examination and intermediated one after radiation treatment in calibrated run. Such samples cannot be classified by the PSL method at all due to the not acceptably low sensitivity. In conclusion, it can be said that reliable classification of the investigated plant extracts by the PSL method based on PN-EN 13751 standard was achieved with 7 samples from among the 16 samples examined. These were: Mulberry extract, Lemon balm extract, Bee balm extract, Celery seed extract, Artichoke extract, Olive extract and Mulberry powder. With the next 7 samples, Asparagus extract, Thyme extract (no. 9, 12, 13 in Table 1), Marigold extract (no. 7 and 10 in Table 1) and Olive leaf extract as well as with 2 other samples showing too low PSL intensity (Melilot extract, Silibina), the classification in PSL is not possible.

In the second stage of the study 36 samples classified earlier by the TL method as non-irradiated have been examined by the PSL method as shown in Table 2.

Screening PSL runs were negative (luminescence intensities below lower threshold value $T_1 = 700$ counts/60 s) with the exception of the sample of Psyllium Compx delivering an intermediate result (714 counts/60 s and 939 counts/60 s, respectively).

Calibrated examination after normalized irradiating of samples with 1 kGy was obtained with 16 samples. These were: Spirulina (no. 2 in Table 2), Citrus aurantium extract, Garlic extract, Bee balm extract, Galanga extract, Dandelion extract (no. 12 in Table 2), Mulberry extract, Celery seed extract, Artichoke extract, Asparagus extract, Citrus aurantium extract, Eyebright extract, Buckwheat extract, Citrus bioflavonoids, Camomile extract and Psyllium Compx. Samples with positive results of calibrated examination, exceeding markedly the negative screening result, can be identified and classified as not irradiated.

With 13 samples tested the calibrated run delivered intermediate results. These were: Thyme extract, Rhodiola rosea extract, Spirulina (no. 5 in Table 2), Ginseng Panax, Grape seed extract, Nettle extract, Dandelion extract (no. 22 in Table 2), Olive leaf extract, Marigold extract, Tribulus terrestris extract, Silybum extract, Valerian extract, Silibina. The negative screening measure and intermediate results of calibrated run show conclusively (see above) that the sample shows low PSL sensitivity. Therefore, such a sample cannot be examined by this method.

The same problem appeared with the next 6 samples that delivered negative results in both screening and calibrated runs (Bilberry extract-two samples, Ginger extract, Schisandra extract, Panax ginseng, Green tea extract). This group of samples is not sensitive to PSL treatment at all.

The classification of 36 samples of plant extracts examined according to PN-EN 13751 standard was possible to be done with 16 samples.

With the next 20 samples the classification was not possible due to the low sensitivity to PSL and the products not sensitive to PSL at all.

Conclusions

In the present study 52 samples of plant extracts have been examined by the PSL method to detect whether irradiated or not.

Two detection methods were applied: thermoluminescence (TL) and photostimulated luminescence (PSL). The reference method was the TL method as the most reliable for the examination of these kinds of foodstuffs. Thus, the reliability of PSL examination was evaluated by a comparison with TL results. The obtained results proved the earlier literature data that

Table 2. Photostimulated luminescence measurements of dry plant extract samples examined by the TL method and identified as non-irradiated

Number of sample	Name of product	Screening PSL	Counts/60 s	Calibrated PSL*	Counts/60 s	Identification of the sample by the PSL method
1	Thyme extract	negative negative	324 304	intermediae intermediae	1703 1072	sample cannot be classified
2	Spirulina**	negative negative	354 317	positive positive	6152 5418	non-irradiated sample
3	Rhodiola rosea extract	negative negative	284 357	intermediae intermediae	1602 916	sample cannot be classified
4	Citrus aurantium extract**	negative negative	476 287	positive positive	34,007 37,738	non-irradiated sample
5	Spirulina**	negative negative	450 263	intermediae intermediae	1395 1262	sample cannot be classified
6	Garlic extract	negative negative	353 289	positive positive	8265 23,626	non-irradiated sample
7	Bilberry extract**	negative negative	379 368	negative negative	488 493	sample cannot be classified
8	Bee balm extract	negative negative	313 261	positive positive	5124 6327	non-irradiated sample
9	Ginger extract	negative negative	242 264	negative negative	409 494	sample cannot be classified
10	Galanga extract	negative negative	311 315	positive positive	5400 7768	non-irradiated sample
11	Schisandra extract	negative negative	360 406	negative negative	556 607	sample cannot be classified
12	Dandelion extract**	negative negative	313 313	positive positive	16,112 8208	non-irradiated sample
13	Mulberry extract**	negative negative	454 447	positive positive	7925 6038	non-irradiated sample
14	Panax ginseng	negative negative	347 358	negative negative	379 440	sample cannot be classified
15	Ginseng Panax	negative negative	532 392	intermediae intermediae	810 765	sample cannot be classified
16	Celery seed extract	negative negative	346 290	positive positive	18,857 23,101	non-irradiated sample
17	Artichoke extract	negative negative	346 419	positive positive	6137 16,161	non-irradiated sample
18	Asparagus extract	negative negative	374 440	positive positive	6259 8769	non-irradiated sample
19	Bilberry extract**	negative negative	480 401	negative negative	580 611	sample cannot be classified
20	Grape seed extract	negative negative	632 608	intermediae intermediae	793 746	sample cannot be classified
21	Nettle extract	negative negative	272 336	intermediae intermediae	2077 1276	sample cannot be classified
22	Dandelion extract**	negative negative	246 306	intermediae intermediae	4303 3596	sample cannot be classified
23	Citrus aurantium extract**	negative negative	233 385	positive positive	12,517 23,088	non-irradiated sample

Table 2. continued.

Number of sample	Name of product	Screening PSL	Counts/60 s	Calibrated PSL	Counts/60 s	Identification of the sample by the PSL method
24	Eyebright extract	negative	372	positive	5582	non-irradiated sample
		negative	330	positive	8901	
25	Olive leaf extract	negative	309	intermediae	1344	sample cannot be classified
		negative	290	intermediae	1696	
26	Marigold extract	negative	270	intermediae	1674	sample cannot be classified
		negative	335	intermediae	1255	
27	Buckwheat extract	negative	408	positive	6799	non-irradiated sample
		negative	343	positive	8576	
28	Citrus bioflavonoids	negative	522	positive	67,259	non-irradiated sample
		negative	433	positive	28,002	
29	Mulberry extract**	negative	411	positive	6092	non-irradiated sample
		negative	379	positive	6703	
30	Green tea extract	negative	296	negative	574	sample cannot be classified
		negative	314	negative	623	
31	Camomile extract	negative	543	positive	15,284	non-irradiated sample
		negative	391	positive	31,421	
32	Tribulus terrestris extract	negative	359	intermediae	1203	sample cannot be classified
		negative	363	intermediae	1011	
33	Silybum extract	negative	365	intermediae	2775	sample cannot be classified
		negative	227	intermediae	2436	
34	Psyllium Compx	intermediate	714	positive	2,388,227	sample cannot be classified
		intermediate	939	positive	1,492,672	
35	Valerian extract	negative	314	intermediae	1451	sample cannot be classified
		negative	316	intermediae	1721	
36	Silibina	negative	374	intermediae	1967	sample cannot be classified
		negative	408	intermediae	1949	

* – after applying 1 kGy normalizing irradiation.
** – samples of the same names but obtained from different sources.

the PSL method, although simple and fast, has limitations arising mainly from the limited PSL sensitivity of some products as observed in this study by examination of plant extracts [1, 5, 7].

Among the 16 samples tested, 7 samples were identified by PSL properly as irradiated. This means that the PSL method was effective roughly in ca. 44% (43.75% as calculated).

In the second PSL study 16 samples identified properly from among 36 as the non-irradiated ones delivered again reliably positive results. The effectiveness of the PSL examination was in this case about 44% too (44.44% as calculated).

The final conclusion of the present investigation is that a fast and relatively simple detection method based on photostimulated luminescence can be only adapted on a limited scale for the detection of radiation treatment of plant extracts. However, the earlier examination of individual extracts by means of both thermoluminescence and photostimulated luminescence is a good proof for further direction of the same kind of sample, not necessarily for thermoluminescence, but perhaps for comparatively reliable in this case examination by pulsed photostimulated luminescence. The construction of such preselection list is the way for faster, i.e. more effective examination of this kind of samples whose preparation in thermoluminescence method meets very often difficulties, too.

References

1. Alberti A, Corda U, Fuochi P, Bortolin E, Calicchia A, Onori S (2007) Light-induced fading of the PSL signal from irradiated herbs and spices. Radiat Phys Chem 76:1455–1458
2. Directive 1999/2/EC (1999) of the European Parliament and of the Council of 22 February 1999; on the approximation of the Member States concerning foods and food ingredients treated with ionising radiation. Off J European Communities L 66/16–23
3. Directive 1999/3/EC (1999) of the European Parliament and of the Council of 22 February 1999; on the establishment of a Community list of food and food ingredients treated with ionising radiation. Off J European Communities L 66/24–25
4. Guzik GP, Stachowicz W (2008) Reliability of light-stimulated photoluminescence (PSL) in detection of irradiated food comparison with thermoluminescence method (TL). Nukleonika 53;S1:s25–s29

5. Malec-Czechowska K, Strzelczak G, Dancewicz AM, Stachowicz W, Delincée H (2003) Detection of irradiation treatment in dried mushrooms by photostimulated luminescence, EPR spectroscopy and thermoluminescence measurements. Eur Food Res Technol 216:157–165
6. PN-EN 1788 (2001) Foodstuffs – thermoluminescence detection of irradiated food from which silicate minerals can be isolated. European Committee for Standardization, Brussels
7. PN-EN 13751 (2002) Foodstuffs – detection of irradiated food using photostimulated luminescence. European Committee for Standardization, Brussels