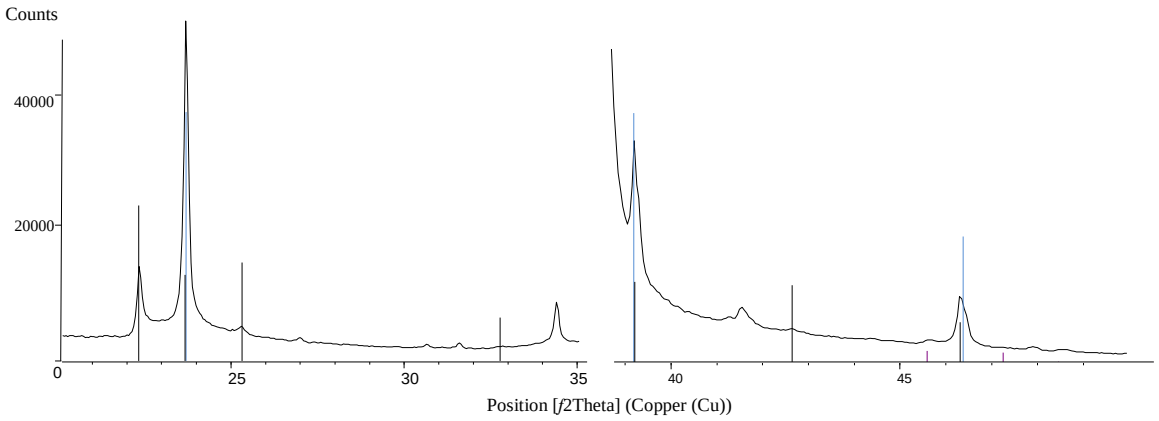
SUPPORTING INFORMATION

A NEW INTEGRATED TLC/MU-ATR/SERS ADVANCED APPROACH FOR THE IDENTIFICATION OF TRACE AMOUNTS OF DYES IN MIXTURES.

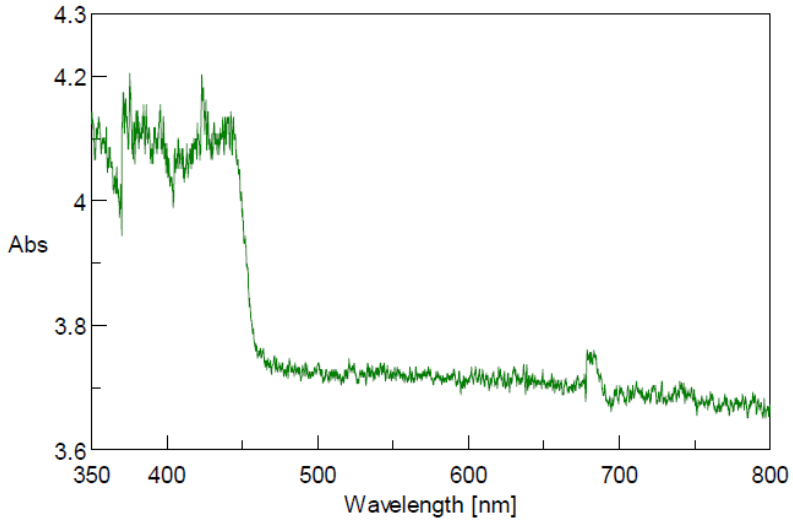
Giorgia Sciutto[1], Silvia Prati[1], Irene Bonacini[1], Lucio Litti[2], Moreno Meneghetti[2], Rocco Mazzeo[1]

1 Microchemestry and Microscopy Art Diagnostic Laboratory, Department of Chemistry, University of Bologna, Via Guaccimannni 42, 48121, Ravenna.

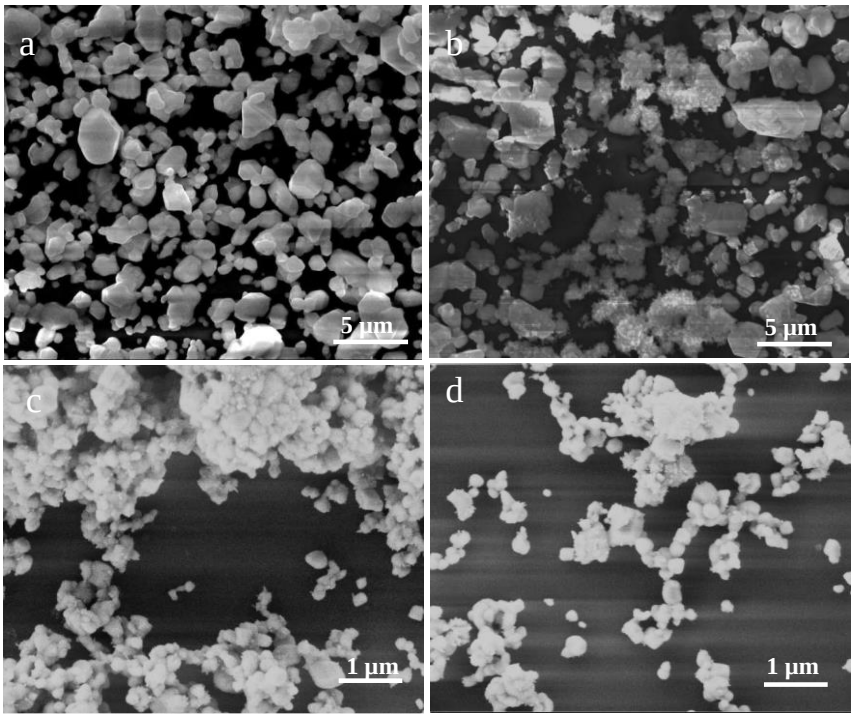
2 Department of Chemical Science, University of Padova, via Marzolo 1, 35131, Padova.



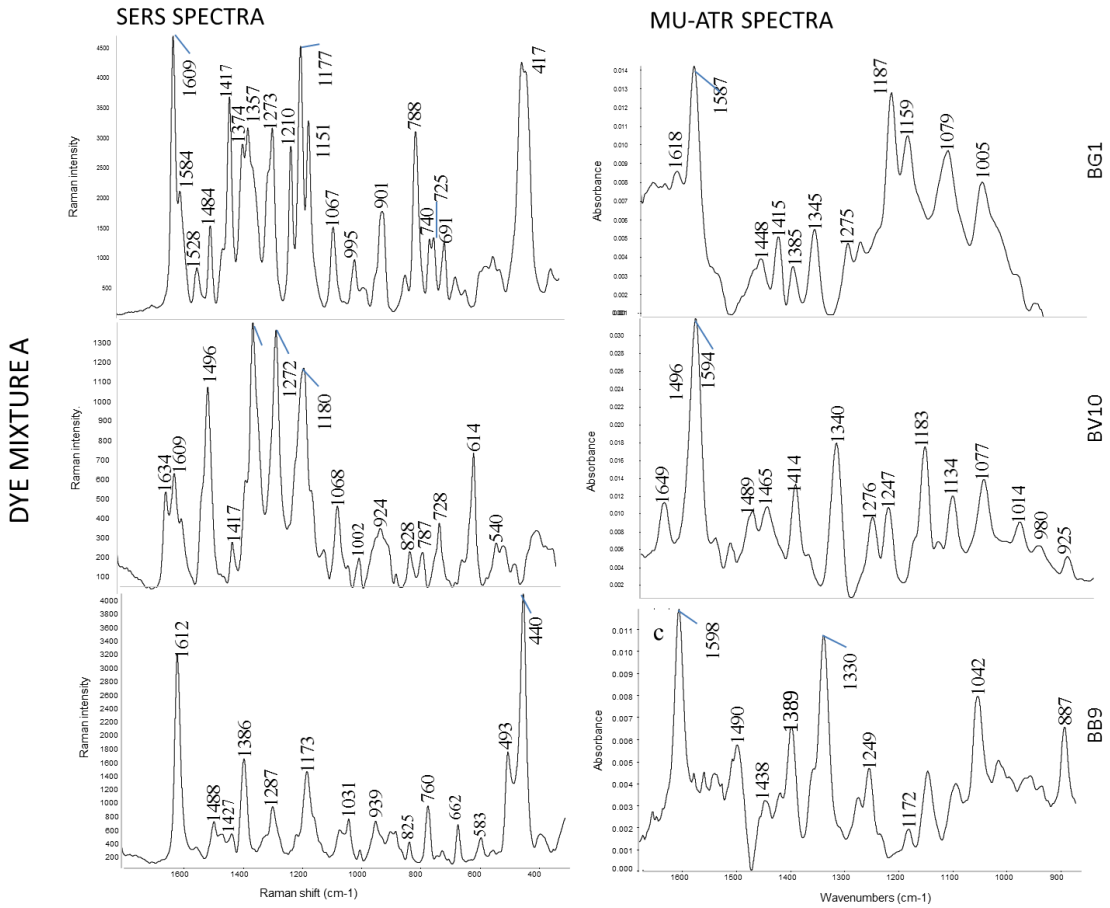
SI Figure 1. XRD diffractogram of AgI particles deposed on an Au substrate.

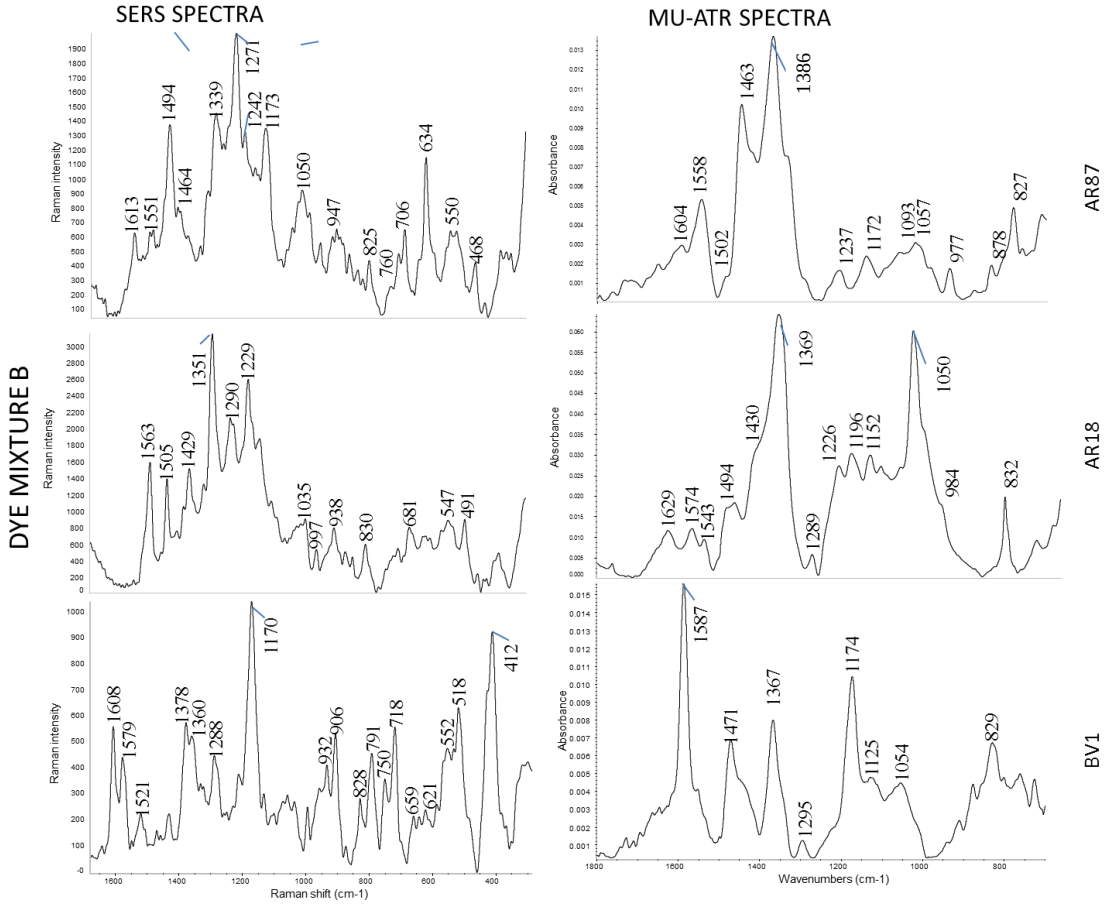


SI Figure 2. UV-Vis spectra of AgI suspension in 2-propanol ([AgI]: 0.014 g/mL)

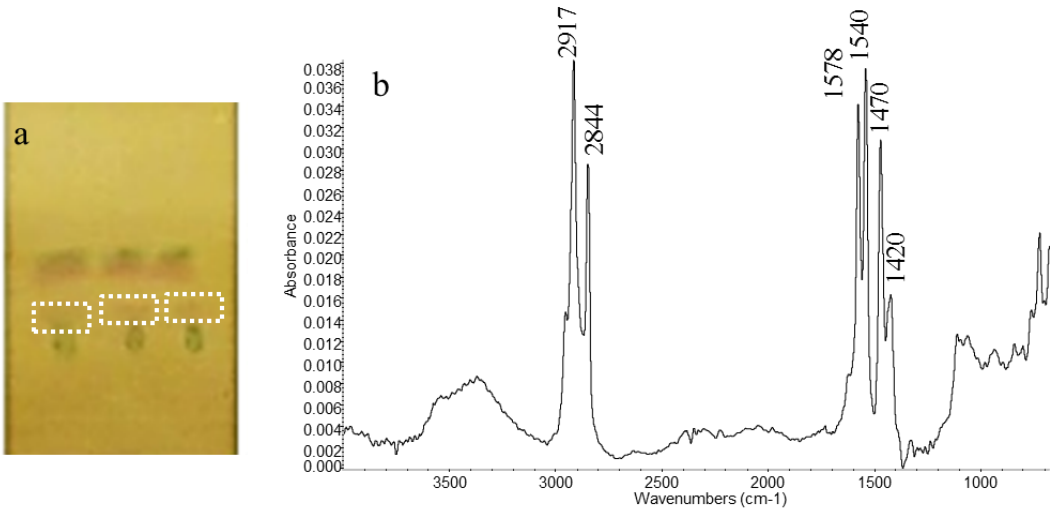


SI Figure 3. SEM images of AgI@SiO_2 substrate before (a, c) and after (b, d) laser irradiation

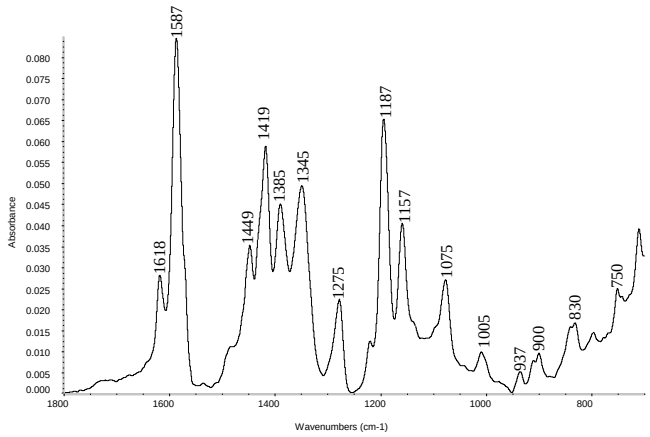

DYE MIXTURE A
SERS SPECTRA
MU-ATR SPECTRA
Raman intensity
Absorbance
Raman shift (cm-1)
Wavenumbers (cm-1)
BG1
BV10
BB9
c

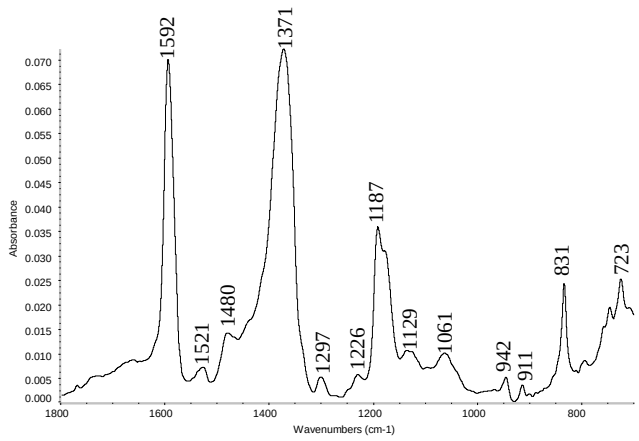
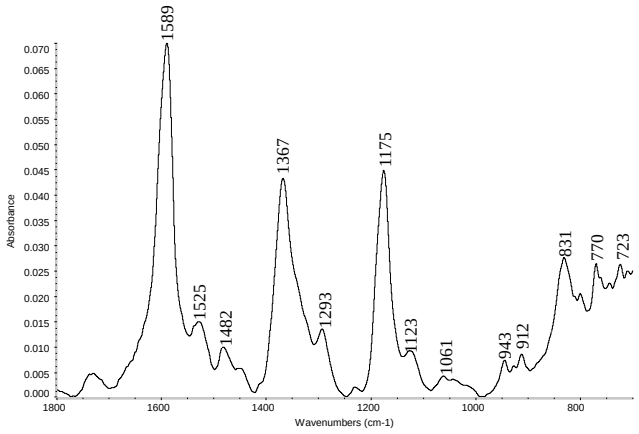
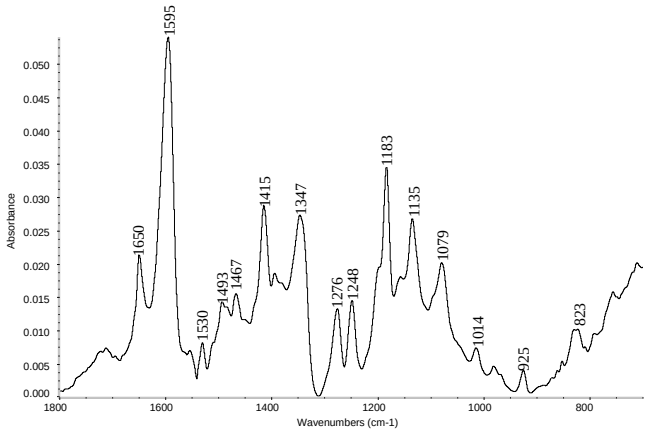

DYE MIXTURE B
SERS SPECTRA
MU-ATR SPECTRA
Raman intensity
Absorbance
Raman shift (cm-1)
Wavenumbers (cm-1)
AR87
AR18
BV1

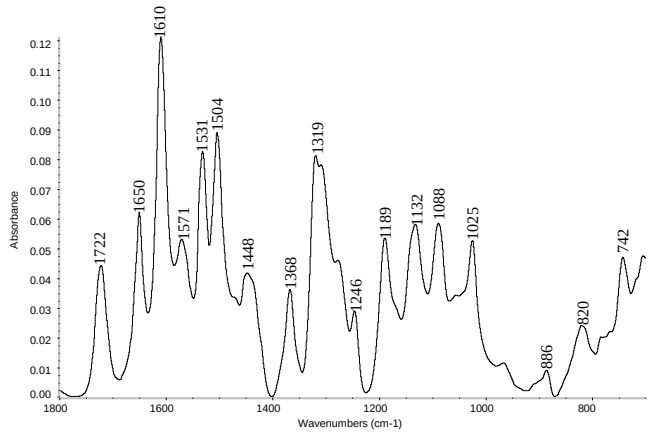
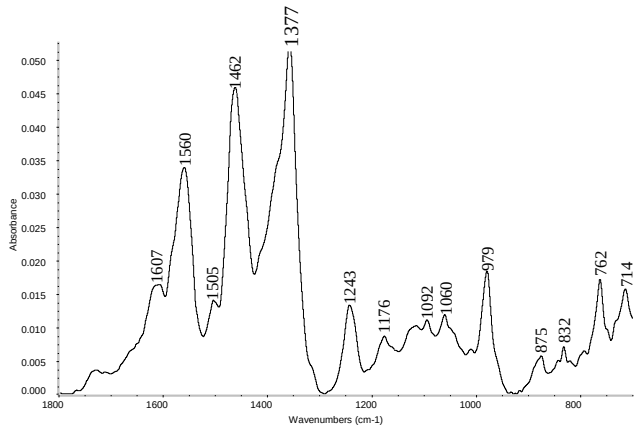
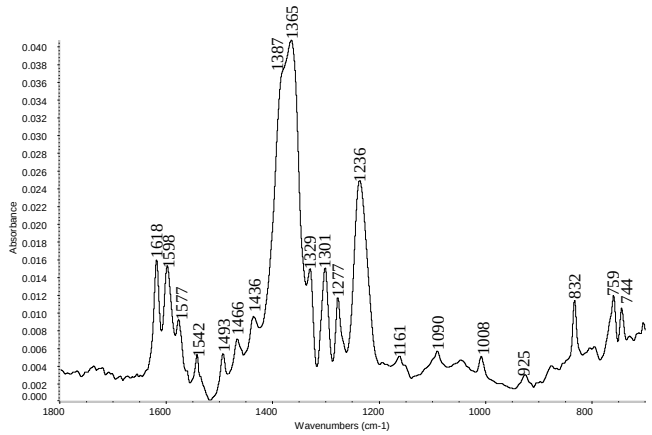
SI Figure 4. SERS and MU-ATR spectra of the dye mixtures A and B after the TLC separation.

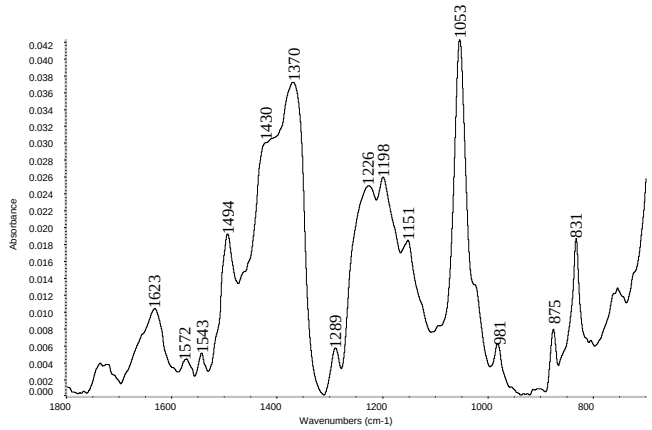
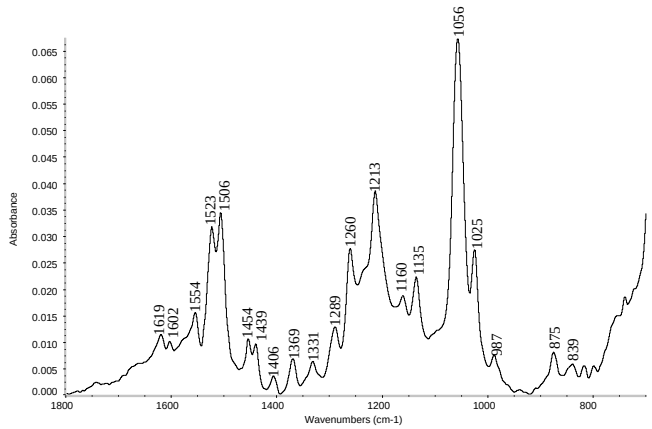
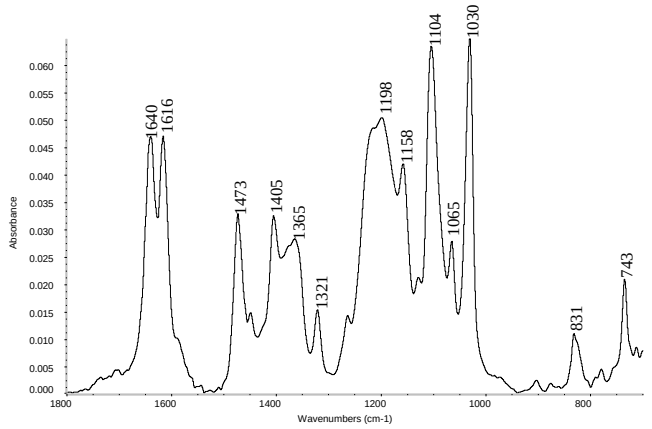


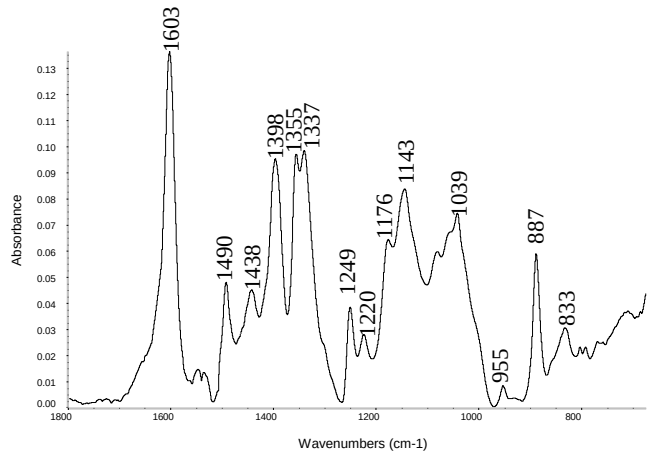
SI Figure 5: a) Dye mixture extracted from the dyed wool after TLC development on AgI@Au plate. White squares indicate areas where the carboxylates have been identified; b) MU-ATR spectrum

Dye Code	**MU-ATR Spectrum on AgI@Au plate**	**Tentative bands assignments**
BG1		1618: aromatic C=C stret.[1]; 1587: C=C str.; 1517: C=C ring str. [1]; 1449: CH_3 def[1]; 1419: CH_2 scissoring def, N-H in plane def; 1385: CH_3 def[1], N-H in plane def. [1]; 1187: C-C str. [1], CH2 wagging[1]; 1157: HSO3- ion[1]; 1075: HSO3- ion S-O sym. Str., C-H in-plane bend.; NH2 rock[1]; 1005: SO3Na sym str[1], asym C-C-N ring bend.[1]

BV3		1592: C=C ring str.[1]; 1521: C=C ring[1] str.; 1480: CH_3 def.; 1297: C-N str.; 1226: Asym C-N str.[1]; 1129: CH in-plane bend[1]; 723: Sym CArCArN str.
BV1		1589: C=C str. ; aromatic C=C str[1]; 1525: C=C str ring[1]; 1482: CH_3 def[1]; 1293: C-N str[1]; 1122: CH in plane bend[1]; 912: Sym. C-N str.[1]; 723: CH3 wagging; C-C out-of-plane bend[1].
BV10		1650:Xanth ring str[2,3]; 1595: Xanth ring str[3]; 1530: Xanth ring -NHC2H5 str[4]; 1347: Xanth. ring C-C str.; 1276: Phen. group[2], C-H bend[3]; 1248: Xanth ring - Phen. [4]; 1183: C-H bend[3]; 1135: Xanth ring -NHC2H5[2]; 1079: Phen ring str[3]; 925: Xanth ring -NHC2H5-phen[2]; 823: Xanth ring -NHC2H5-phen[2].

BR1		1722: C=O str. [4]; 1650: Xanth ring str.[4]; 1610: Xanth ring str; 1570 phen. str. [4]; 1448: C=O str. [4]; 1368: COO^- str.; 1319: Xanth. ring CC str.; 1189: Xanth ring str. [4].
AR87		1607: Xanth ring str.[6,7]; 1560: Xanth ring str.[6]; 1462: C=O str.[7]; 1377: COO^- str.; 1243: C-H in-plane bend[6]; 979: C-H out-of-plane bend[6,7]; 762: C-H out-of-plane bend[6,7]; 714: C-H out-of-plane bend[6,7].
AY2 [8]		1618: aromatic C=C str, C-N bend; 1598: NO2 rock., aromatic C=C str; 1577: NO2 sym str.; 1496: aromatic C-C str.; 1387: aromatic C-C str., NO2 sym str., 1329: NO2 sym str, NO2 sym bend., NO2 rock., C-N str.; 1301: RC-H bend, aromatic C-C str.; 1277: RC-H bend, R1 trigonal def.; 1240: RC-H bend, C-O bend.; 759: R1 asym. torsion, C-H wag, R1 puckering, CN wagging; 744: C-N bend., aromatic C-C str., NO2 sym bend., C-O bend.

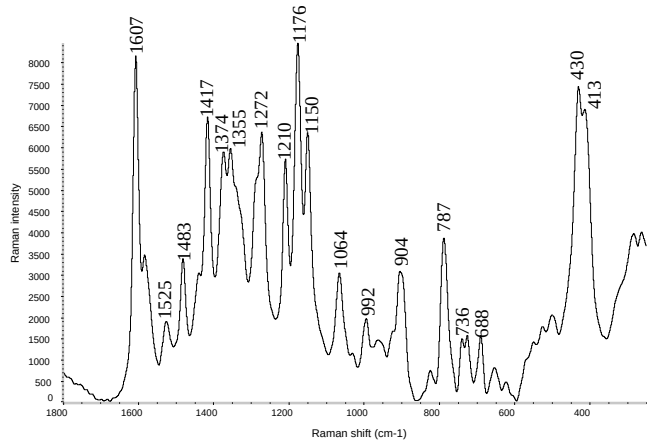
AR18 [9]		1543: N-H bend., C=N str., C-C str., N-H bend.; 1494: N-H bend., C-O str., C=N str.; 1430: N=N str.; 1370: C-C str., C-N str., 1289: C-N str, C-H in-plane bend.; 1198: Asym. str., SO3-, C-S str.; 1151: C-S, SO3; 1053: Sym. Str. SO3-, C-S str.; 831: C-H out-of-plane def.
AR88		1619: N=N str[10]; 1523: aromatic C=C str. [10]; 1506: aromatic C=C str. [10]; 1368: N=N str.[11]; 1160: C-O str.; 1213: C-O str.; 1056: S=O str. [11]; 839: C-H out-of plane bend. [10]
AB74		1640: aromatic C=C str. [12]; 1615: C=C str.; [12, 13]; 1104: asym. benzenesulfonic acids str. [13]; 1030 SO3 str; [12].

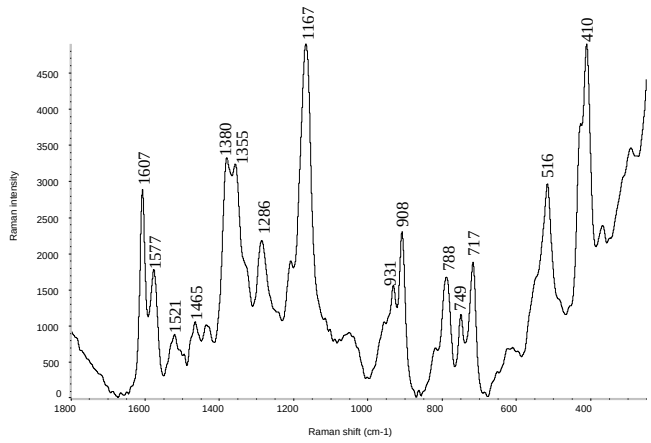
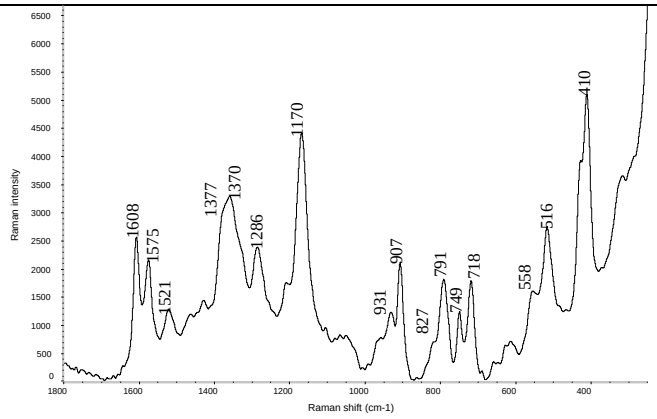
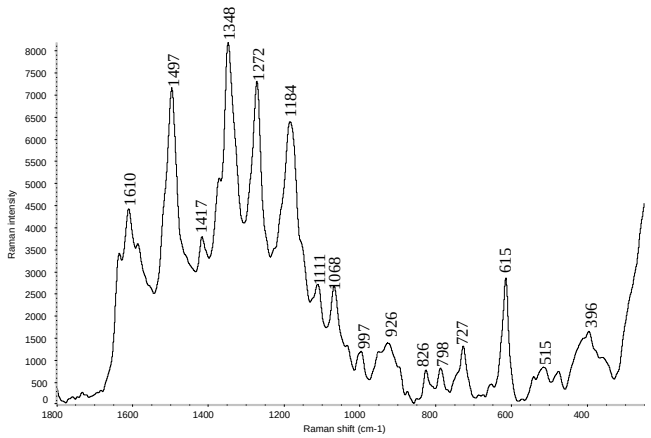
BB9		1603: Aromatic C=C str.[14]; antisym. C-N str.[15]; C=C str. [13]; C-H in-plane bend. [15]; 1490: C=N str.; C=C str.; C-H out of plane bend. [15]; 1438: Asym. C-H bend.[16]; antisym. C-N str. [15]; C-H in-plane bend. [15]; out-of-plane C-H bend. [15]; 1398: sym. C-H bend.; sym. C-H bend. [16]; antisym. C-N str. [16]; C-H in-plane bend. [16]; C-H out-of-plane bend. [16]; 1355: sym. Ar-N str. [14]; C-H in-plane bend. [15]; C-H out-of-plane bend. [15]; 1249: C-H in-plane bend. [16]; 1220: antisym. C-N str. [15]; C-H in plane bend. [16]; C-H out-of-plane bend. [15]; 1176: C-H out-of-plane bend. [16]; C-H in-plane bend. [15]; 1143: C-H in plane bend. [15]; 1039: antisym. C-S str. [15]; C-H in plane bend. [15]; 887: C-H bend. [15]; C-H out of plane bend. [15]

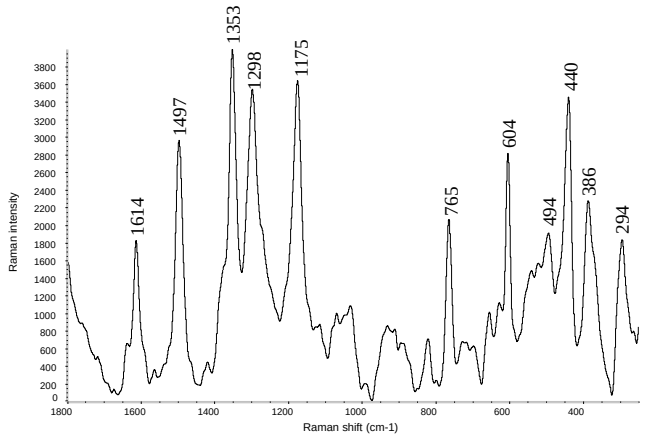
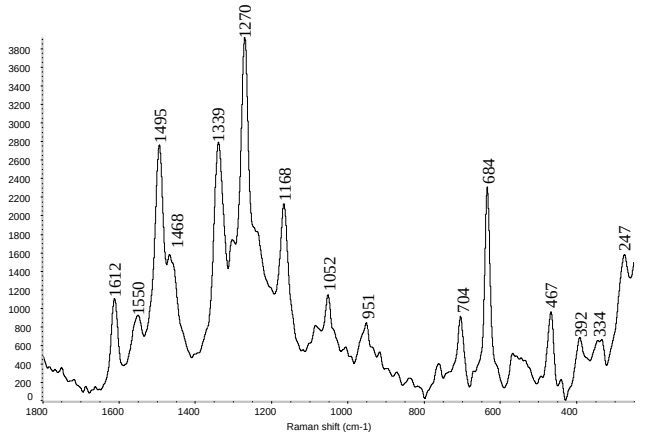
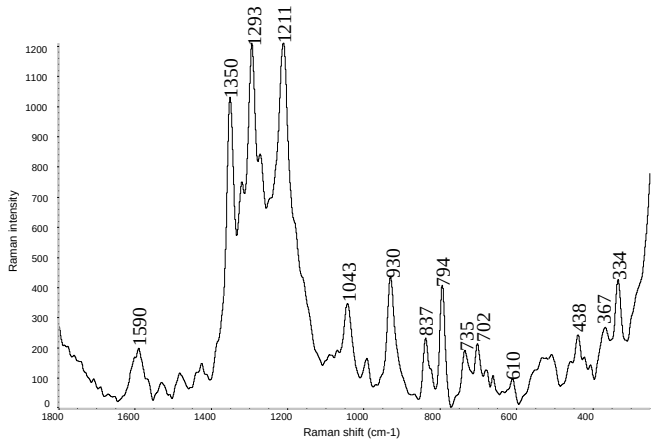
SI Table 1. MU-ATR spectra of the investigated synthetic dyes on AgI@Au TLC plates

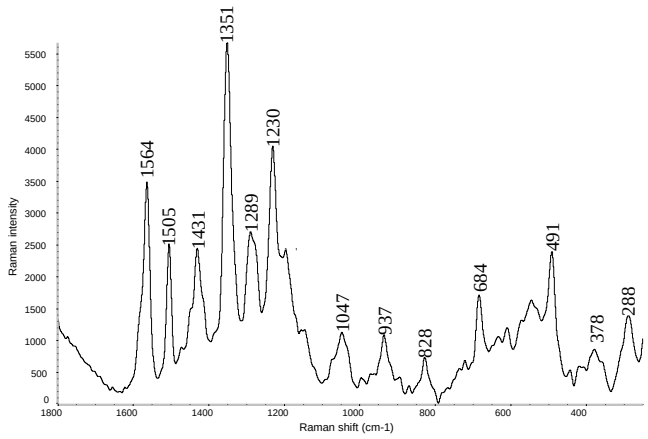
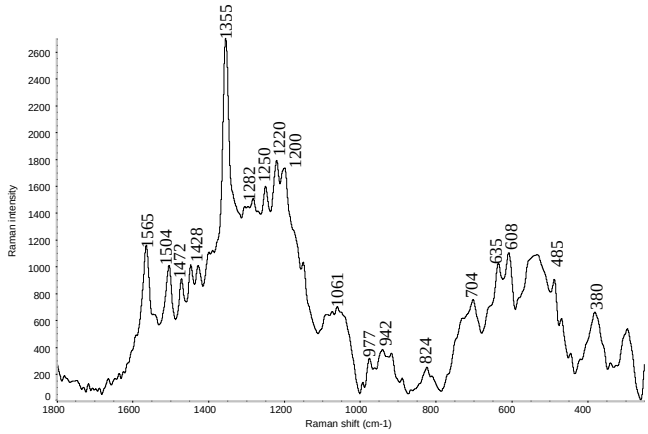
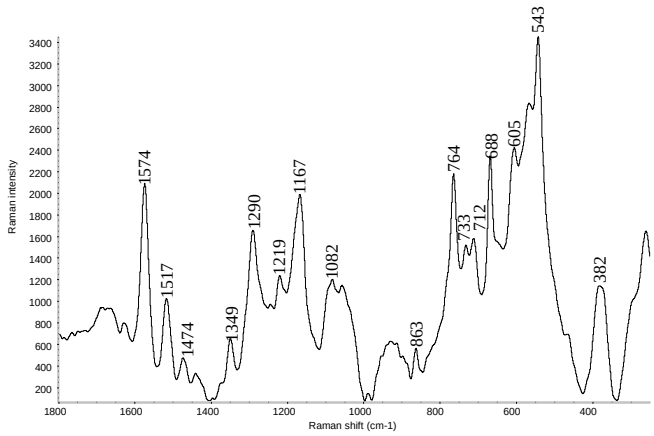
1. Doherty, B., Vagnini, M., Dufourmantelle, K., Sgamellotti, A., Brunetti, B., & Miliani, C. (2014). A vibrational spectroscopic and principal component analysis of triarylmethane dyes by comparative laboratory and portable instrumentation. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, *121*, 292-305.
2. Sharma A., Kumar A., Vibrational (2013)study of Rhodamine 610 and effect of methanol on its vibrational spectra; Greener Journal of Physical Sciences *3*(6), 200-203.
3. Sarkar, J., Chowdhury, J., Pal, P., & Talapatra, G. B. (2006). Ab initio, DFT vibrational calculations and SERRS study of Rhodamine 123 adsorbed on colloidal silver particles. *Vibrational spectroscopy*, *41*(1), 90-96.
4. Watanabe, H., Hayazawa, N., Inouye, Y., & Kawata, S. (2005). DFT vibrational calculations of rhodamine 6G adsorbed on silver: analysis of tip-enhanced Raman spectroscopy. *The Journal of Physical Chemistry B*, *109*(11), 5012-5020.
5. Narayanan, V. A., Stokes, D. L., & Vo-Dinh, T. (1994). Vibrational spectral analysis of Eosin Y and Erythrosin B—intensity studies for quantitative detection of the dyes. *Journal of Raman spectroscopy*, *25*(6), 415-422.
6. Hazebroucq, S., Labat, F., Lincot, D., & Adamo, C. (2008). Theoretical insights on the electronic properties of eosin Y, an organic dye for photovoltaic applications. *The Journal of Physical Chemistry A*, *112*(31), 7264-7270.
7. Markuszewski, R., & Diehl, H. (1980). The infrared spectra and structures of the three solid forms of fluorescein and related compounds. *Talanta*, *27*(11), 937-946.
8. Snehalatha, M., Ravikumar, C., Joe, I. H., & Jayakumar, V. S. (2009). Vibrational spectra and scaled quantum chemical studies of the structure of Martius yellow sodium salt monohydrate. *Journal of Raman Spectroscopy*, *40*(9), 1121-1126.

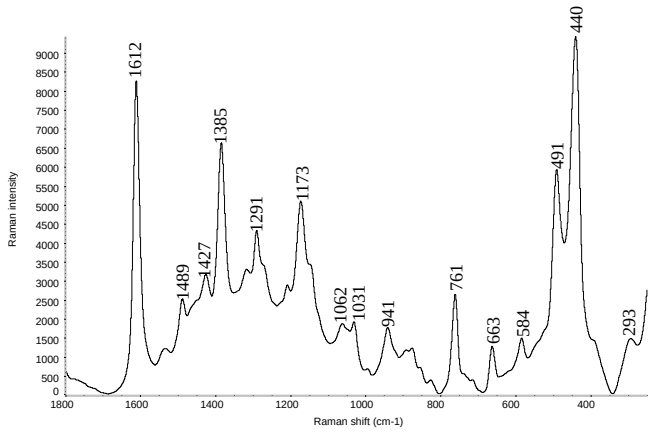
9. Almeida, M. R., Stephani, R., Dos Santos, H. F., & Oliveira, L. F. C. D. (2009). Spectroscopic and theoretical study of the "azo"-dye E124 in condensate phase: evidence of a dominant hydrazo form. *The Journal of Physical Chemistry A*, *114*(1), 526-534.
10. Dikmen, S., Gunay, A., Ersoy, B., & Erol, I. (2015). DETERMINATION OF THE EQUILIBRIUM, KINETIC AND THERMODYNAMIC PARAMETERS OF ACID RED 88 ADSORPTION ONTO MONTMORILLONITIC CLAY. *Environmental Engineering and Management Journal*, *14*(5), 1097-1110.
11. Anil Kumar, M., Vinoth Kumar, V., Ponnusamy, R., Paul Daniel, F., Seenuvasan, M., Anuradha, D., & Sivanesan, S. (2015). Concomitant mineralization and detoxification of acid red 88 by an indigenous acclimated mixed culture. *Environmental Progress & Sustainable Energy*, *34*(5), 1455-1466.
12. Vautier, M., Guillard, C., & Herrmann, J. M. (2001). Photocatalytic degradation of dyes in water: case study of indigo and of indigo carmine. *Journal of Catalysis*, *201*(1), 46-59.
13. Galindo, C., Jacques, P., & Kalt, A. (2001). Photochemical and photocatalytic degradation of an indigoid dye: a case study of acid blue 74 (AB74). *Journal of Photochemistry and Photobiology A: Chemistry*, *141*(1), 47-56.
14. Sáez, E. I., & Corn, R. M. (1993). In situ polarization modulation—Fourier transform infrared spectroelectrochemistry of phenazine and phenothiazine dye films at polycrystalline gold electrodes. *Electrochimica acta*, *38*(12), 1619-1625.
15. Aoki, P. H., Volpati, D., Caetano, W., & Constantino, C. J. (2010). Study of the interaction between cardiolipin bilayers and methylene blue in polymer-based Layer-by-Layer and Langmuir films applied as membrane mimetic systems. *Vibrational Spectroscopy*, *54*(2), 93-102.
16. Ovchinnikov, O. V., Chernykh, S. V., Smirnov, M. S., Alpatova, D. V., Vorob'Eva, R. P., Latyshev, A. N., ... & Lukin, A. N. (2007). Analysis of interaction between the organic dye methylene blue and the surface of AgCl (I) microcrystals. *Journal of Applied Spectroscopy*, *74*(6), 809-816.

Dye Code	SERS-Raman Spectrum on AgI@Au plate	Tentative bands assignments
BG1		1607: aromatic C-C str. [1]; 1528: C-N str.; sym. CH3 bend.[1]; 1481: asym. C-H bend.[1]; 1374: C-C str. [1]; 1357: asym. C-C str.; C-C bend.; C-H bend.[1]; 1272: asym. C-C str.; C-C bend.[2]; C-H bend. [1]; 1210: sym. C-C str.; C-C bend.; C-H bend. [2]; 1176: C-H bend. [1]; 1150: sym. C-C str.; C-C bend.; C-H bend. [1]; 995: C-C bend. [2]; 904: C-C str.[1]; 788: C-C str.; C-N str.[1]; 736: C-N str.[1]; 430: C-N bend.; 414: CNC bend.; C-C bend., SO3

		bend. [2]
BV3[1]		1607: aromatic C-C str.; 1577: aromatic C-C str.; 1380: C-C str.; 1355: asym. C-C str., C-C-C ring bend.; C-H bend.; 1171: sym. C-C str.; C-C-C bend.; C-H bend.; 908: C-C ring str.; 717: C-N str.; 410:C-N-C bend.; C-C-C bend.
BV1[1]		1584: aromatic C-C str.; 1170: sym. C-C-C str.; C-C-C bend.; C-H bend; 907: C-C ring str.; 797: sym. C-C-C str.; C-N str.;
BV10		1610: Xhant. ring str.[3]; 1497: Xhant. ring str. [3]; 1348: Xhant. ring str. [3]; 1272: C-H bend. [3]; 1180: C-H bend.; 924: Xhant. ring + phenyl str.; 727: C-H out-of-plane bend. of phenyl ring.; 615: Xanth. ring in.plane def.[4]

BR1[4]		1497: Xanth. ring str.; C-N str.; C-H bend.; N-H bend.; 1353: Xanth. Ring str.; C-H in-plane bend.; 1298: Xanth. Ring def.; N-H bend.; CH2 wagging.; 1175: Xanth. ring in-plane def.; C-H bend.; N-H bend.; 765: Xanth. ring in-plane def.; 604: Xanth. ring in-plane def.
AR87[5]		1612: Xanth. C-C ring str.; 1495: Xanth. C-C ring str.; 1339 Xanth. C-C ring str.; 1270: C-C str.; 1052: C–C str.; sym C-O str.; 1270: Xanth. and Bz C-C ring str.1052: Bz ring str.; sym. CO2 str.; 704: Xanth. out-of-plane ring def.
AY2[6]		1211: Naphthalene C–H bend. ; 1143: C–H out-of-plane bend. 1350: C-C str.; 930: NO_2 def.; 702: NO_2 def.

AR18[7]		1564: aromatic C=C str.; N-H bend.; 1503: C=O str., C=N str.; N-H bend.; 1431: C-H str.; 1351: C-C str.; 1289: N-N str.; C-C str.; 1230: C-C str.; N-N str.; 491: out-of-plane ring def.
AR88[8]		1565: aromatic C=C str.; 1355: N=N str.; 1200: sym. C-N str.
AB74[9]		1584: Asym. C=C str.; C=O str.; C=N str.; 1474: C-C str.; C-H bend.; C=N str.; 1290: sym. SO3 str.; C-C bend.; 1082: O-Na bend.; C-H bend.; C-C bend.; 863: N-H bend.; C-N str.; 543: (C=C-CO-C) bend.

BB9[10]		1612: C-C str.; C-H in-plane bend.; 1489: sym. C-N str.; C-H in-plane bend.; C-H out-of-plane bend.; 1427: C-N str C-H in-plane bend.; C-H out-of-plane bend.; 1385: C-N str C-H in-plane bend.; C-H out-of-plane bend.; 1173: -H in-plane bend.; C-H out-of-plane bend.; 761: skeletal deformation (CN and CH_3); 662: skeletal deformation (CC); 491: skeletal deformation (CN, CS and CH_3);

SI Table 2. Raman spectra of the investigated synthetic dyes on AgI@Au TLC plate

1. Doherty, B., Vagnini, M., Dufourmantelle, K., Sgamellotti, A., Brunetti, B., & Miliani, C. (2014). A vibrational spectroscopic and principal component analysis of triarylmethane dyes by comparative laboratory and portable instrumentation. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, *121*, 292-305.
2. Wu, M. C., Lin, M. P., Chen, S. W., Lee, P. H., Li, J. H., & Su, W. F. (2014). Surface-enhanced Raman scattering substrate based on a Ag coated monolayer array of SiO 2 spheres for organic dye detection. *RSC Advances*, *4*(20), 10043-10050.
3. Sarkar, J., Chowdhury, J., Pal, P., & Talapatra, G. B. (2006). Ab initio, DFT vibrational calculations and SERRS study of Rhodamine 123 adsorbed on colloidal silver particles. *Vibrational spectroscopy*, *41*(1), 90-96.
4. Jensen, L., & Schatz, G. C. (2006). Resonance Raman scattering of rhodamine 6G as calculated using time-dependent density functional theory. The Journal of Physical Chemistry A, 110(18), 5973-5977.
5. Greeneltch, N. G., Davis, A. S., Valley, N. A., Casadio, F., Schatz, G. C., Van Duyne, R. P., & Shah, N. C. (2012). Near-infrared surface-enhanced Raman spectroscopy (NIR-SERS) for the identification of Eosin Y: theoretical calculations and evaluation of two different nanoplasmonic substrates. *The Journal of Physical Chemistry A*, *116*(48), 11863-11869.
6. Snehalatha, M., Ravikumar, C., Joe, I. H., & Jayakumar, V. S. (2009). Vibrational spectra and scaled quantum chemical studies of the structure of Martius yellow sodium salt monohydrate. Journal of Raman Spectroscopy, *40*(9), 1121-1126.
7. Almeida M.R., (2010) Spectroscopic and Theoretical Study of the "Azo"-Dye E124 in Condensate Phase: Evidence of a Dominant Hydrazo Form, The Journal of Physical Chemistry A *114,* 526-534.
8. Schulte, F., Brzezinka, K. W., Lutzenberger, K., Stege, H., & Panne, U. (2008). Raman spectroscopy of synthetic organic pigments used in 20th century works of art. *Journal of Raman spectroscopy*, *39*(10), 1455-1463.
9. Peica, N., & Kiefer, W. (2008). Characterization of indigo carmine with surface-enhanced resonance Raman spectroscopy (SERRS) using silver colloids and island films, and theoretical calculations. *Journal of Raman Spectroscopy*, *39*(1), 47-60.
10. Aoki, P. H., Volpati, D., Caetano, W., & Constantino, C. J. (2010). Study of the interaction between cardiolipin bilayers and methylene blue in polymer-based Layer-by-Layer and Langmuir films applied as membrane mimetic systems. *Vibrational Spectroscopy*, *54*(2), 93-102.