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LC-TOF/MS Analysis for Halal Identification



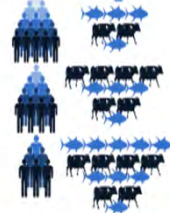
Disampaikan dalam Kajian Halal & Pembukaan Seminar Nasional Safety dan Halal Undip 2018



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?????



Peningkatan Populasi

➔

Intensifikasi Produksi



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Food Safety

– dalam arti luas mencakup :

- Tidak hanya mengenai efek akut namun juga kronis : karsinogen, teratogenik, mutagenic, dll
- Mempertimbangkan dampak terhadap ekologi, lingkungan dan makhluk hidup lainnya
- Labelling dan hak konsumen untuk memilih : alergen, organik, GMO, halal



Akumulasi senyawa karsinogen pada manusia bisa menyebabkan penyakit kanker (penyakit kronis)



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Food Safety

MANDATORY !!

Spesifikasi Standar :

- Batasan cemaran mikroba
- Batasan cemaran ogam berat
- Batasan cemaran residu pestisida
- Batas penambahan pengawet, penyedap buatan dan pewarna, dll.

Food Quality

faktor eksternal seperti penampilan (ukuran, bentuk, warna dan konsistensi), tekstur, rasa; dan *product traceability*

Voluntary



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Food Authenticity

- Allergen
Crustacea, telur, ikan, Lupin, Sereal mengandung gluten, Susu (termasuk laktosa), Moluska, Mustard, Kacang, Kacang Tanah, biji wijen, Kedelai, SO₂ (Sulfit) dan Seledri
- Keaslian dari komoditas, contoh :
Wine, Olive oil, kopi and chocolate, ikan kakap merah




Figure 1: The "Big Eight" Allergens: Soy, Milk, Peanuts, Egg, Milk, Fish, Wheat and Shellfish.

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FOOD INTEGRITY

Quality Safety Authenticity

Consumer expectations

- Expect **safety, quality** and **authenticity** to be assured
- Accurate labelling "It does what it says on the can"
- The Food Information for Consumers Regulation –embraces this sentiment
- They expect **food integrity** to be assured

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FOOD INTEGRITY: a comprehensive concept



Primary Production Storage Processing Packaging Distribution Retail Product Consumption

FOOD INTEGRITY

Verification procedures

Supply Chain Management, Risk management procedures

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Yang harus diperhatikan:

! tidak sengaja / poor management

- Kesalahan memilih Lahan → Tercemar Logam Berat
- Bad Agricultural Practice → Mengandung pestisida
- Kesalahan paska panen → Tercemar Mycotoxin
- Kesalahan Proses Produksi → Tercemar PAH's

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! Yang harus diperhatikan:

- Faktor kesengajaan

The diagram shows four interlocking gears of different colors: a purple gear labeled 'Misused' at the top, a green gear labeled 'Adulteration' on the left, a red gear labeled 'Fraud' on the right, and a blue gear labeled 'Crime' at the bottom. Arrows indicate a clockwise flow between the gears.

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- Faktor kesengajaan

Misused (penyalahgunaan)

The top right shows a close-up of a fish's head and a pile of yellow pills, with the text 'chemaphermed' below the pills.

Three panels are shown in a row, each with a different image: a goldfish, a water filter, and a woman. A large blue arrow points from right to left across the bottom of these panels, with the text 'Tidak sama peruntukannya' (Not the same for use) written inside it.

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Adulteration (pemalsuan)

A plate of golden-brown chicken nuggets with a small garnish of green herbs.

Nugget Ayam Original

Berapa % kandungan ayam dibanding dengan bahan tambahan lain

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Fraud (penipuan)

A collection of various animals including a horse, a rabbit, a shrimp, a fish, a chicken, a dog, a pig, a mouse, a goat, and a cow. In the center is a blue starburst containing the text 'FOOD AUTHENTICITY LABEL' above a can with the word 'LABEL' written vertically on it.

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Fraud (penipuan)

Seberapa % tingkat keasliannya

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Food Crime

Motivasi keuntungan

Melamine + pakan ikan = Susu bayi

seolah-olah kadar protein tinggi

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Perkembangan teknologi pengujian

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Target Concentration

mg/kg (ppm)
ug/kg (ppb)
ng/kg (ppt)
pg/kg (ppq)

How Low You Go?

Metric prefixes							
Prefix	Symbol	1000 th	10 th	Decimal	Short scale	Long scale	Since ⁽ⁿ⁻¹⁾
		1000 ⁰	10 ⁰	1	one	-	
deci	d	1000 ^{-1/3}	10 ⁻¹	0.1	tenth	1795	
centi	c	1000 ^{-2/3}	10 ⁻²	0.01	hundredth	1795	
milli	m	1000 ⁻¹	10 ⁻³	0.001	thousandth	1795	
micro	μ	1000 ⁻²	10 ⁻⁶	0.000 001	millionth	1960	
nano	n	1000 ⁻³	10 ⁻⁹	0.000 000 001	billionth	milliardth	1960
pico	p	1000 ⁻⁴	10 ⁻¹²	0.000 000 000 001	trillionth	billionth	1960
femto	f	1000 ⁻⁵	10 ⁻¹⁵	0.000 000 000 000 001	quadrillionth	billiardth	1964
atto	a	1000 ⁻⁶	10 ⁻¹⁸	0.000 000 000 000 000 001	quintillionth	trillionth	1964
zepto	z	1000 ⁻⁷	10 ⁻²¹	0.000 000 000 000 000 000 001	sextillionth	trilliardth	1991
yocto	y	1000 ⁻⁸	10 ⁻²⁴	0.000 000 000 000 000 000 000 001	septillionth	quadrillionth	1991

1. * The metric system was introduced in 1795 with subprefixes. The other dates in this table are given by a convention of the CIPM.

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International Trend

- > Negative List ⇒ Positive list (Target Analytes ↑)
- > Targeted ⇒ + Non-Targeted Screening

“Measuring” change - techniques

Non-targeted

Increasing specificity ↓

FT-IR spectroscopy

NMR spectroscopy

Mass spectrometry

↑ Increasing coverage

Targeted

EVOLUTION

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INSTRUMENT SELECTION

Based on Commission Decision 2002/657/EC or screening and confirmatory analysis. (Vet drugs, Pesticides, Aflatoxins, Elements)

Increasing potential for interferences

Increasing ability to discriminate

Simple Extraction

Suitable for use as confirmatory assays for BANNED compounds

Suitable for use as confirmatory assays for MRL compounds

Suitable for use as screening assays

TOF/MS
MS/MS
LC/GC-MS
HPLC-F
HPLC-UV
ELISA
Microbiological

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APA SAJA YANG BISA DI UJI DENGAN MS UNTUK HALAL

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HALAL

- SYARIAH (muslim only)
 - HALAALAN
 - Qualitative: Porcine Screening (DNA: PCR, Polypeptide: LC-MS/MS, Lipid: LC-MS/MS, FAME: GCMS)
 - Quantitative: Alcohol Limit Screening (Ethanol: GCMS)
- SAFETY (muslim & Non Muslim)
 - TOYYIBAN
 - Qualitative: Adulterant Screening (for men health product) (Sildenafil, tadalafil, Vardenafil: LCMS)
 - Steroid screening (for cosmetic product) (Depend to type of cosmetic range of product)
 - Quantitative: Heavy metal limit test (Pb, Cd, Hg, As: ICP-MS), Microbial limit test (Total aerobic microbial count, Total Yeast & Mold)

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Akurasi identifikasi halal dengan MS

Sebagai contoh perbandingan dalam pengujian porcine dengan PCR

Purity of DNA

A_{260}/A_{280} :
dsDNA = 1.8
pure RNA = 2.0
Protein <1, 0.5 or so

Pengukuran Rasio absorbansi pada 260 nm dan 280 nm digunakan untuk menilai kemurnian dari DNA dan RNA.

FALSE POSITIVE **FALSE NEGATIVE**

MS
Phenotype/Function

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MS Popularity

Environmental, Health-Related and Economic Factors

- Need to Assess Risks for Humans and Environment
- Need to Improve Productivity and Sample Throughput & to Reduce TAT and Costs
- Need to Reduce Solvent Consumption

Advancements in Instrumentation (Electronics, Robotics, IT...)

GC/ITD, MSD, MS-MS, TOF, PTV
LC/MS, MS-MS, TOF
Selective detectors ECD, NPD, FPD

1960 1970 1980 1990 2000 2010

MS technology is popular today

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Kemudahan uji dengan menggunakan MS

1. High End Equipment
2. High Throughput
3. Simple Preparation
4. General Scope

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Tipe sampel yang dapat diuji

- Makanan
- Pakan
- Pupuk
- Sampel Lingkungan
- Farmasi

Single / Multi / Mix

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Instrument **UPLC-UHR-Qq-TOF-MS**

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Resolution

Res. 1'000

MW 1672.9

Res. 40'000

Resolution = (m/z) / FWHM

23-Jul-18

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Isotope pattern

- Intensity of all isotopes
- Mass position (distance) of all isotopes

$^{13}\text{C} - ^{12}\text{C} : 1.0033548 \text{ Da}$

$^2\text{H} - ^1\text{H} : 1.00627675 \text{ Da}$

$^{15}\text{N} - ^{14}\text{N} : 0.997035 \text{ Da}$

$^{34}\text{S} - ^{32}\text{S} : 1.9957959 \text{ Da}$

simulated spectrum of reserpine $[\text{M}+\text{H}]^+ \text{C}_{33}\text{H}_{41}\text{N}_2\text{O}_9$

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Identification based on accurate mass and true isotopic pattern in MS


Meas. m/z	#	Formula	m/z	err [Da]	err [ppm]	nSigs	# Sigs
416.3160	1	C ₂₆ H ₄₂ N ₂ O ₃	416.3159	-0.1	-0.2	5.2	1
417.3188	2	C ₂₆ H ₄₂ N ₂ O ₃	417.3185	0.3	1.2	21.1	2

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Analisis dapat dilakukan di TOF-MS

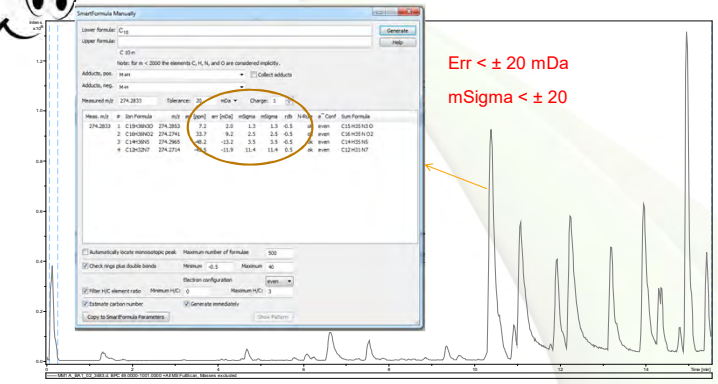
1. Qualitative Screening Non Targeted → Data Analysis
2. Qualitative Screening Targeted → Target Analysis
3. Quantitative Screening Targeted → Quant Analysis
4. Profiling → Profile Analysis



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Qualitative Screening Non Targeted

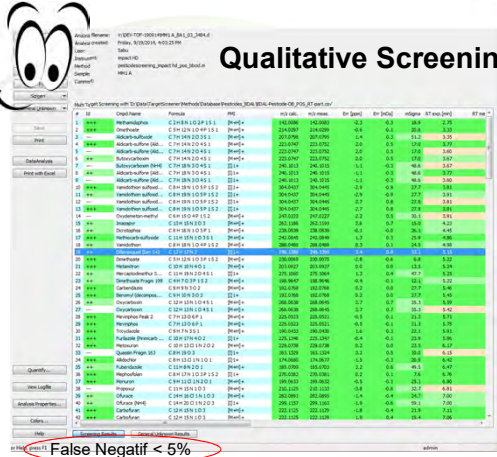


Err < ± 20 mDa
mSigma < ± 20

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Qualitative Screening Targeted



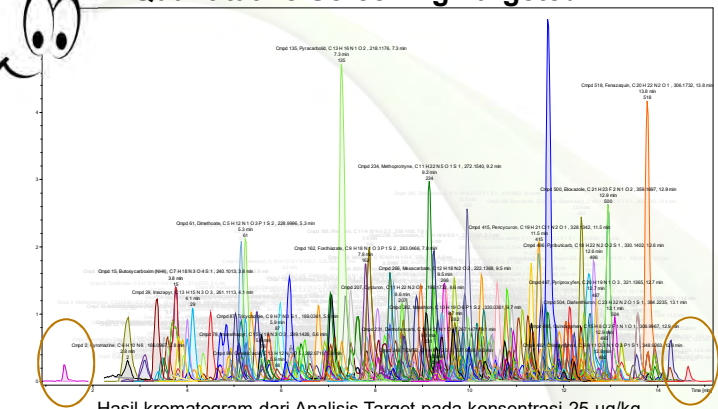
Results of the assessment:
+++ : very good
++ : good
- : large deviations for RT, mass or sigma results require additional information

False Negatif < 5%

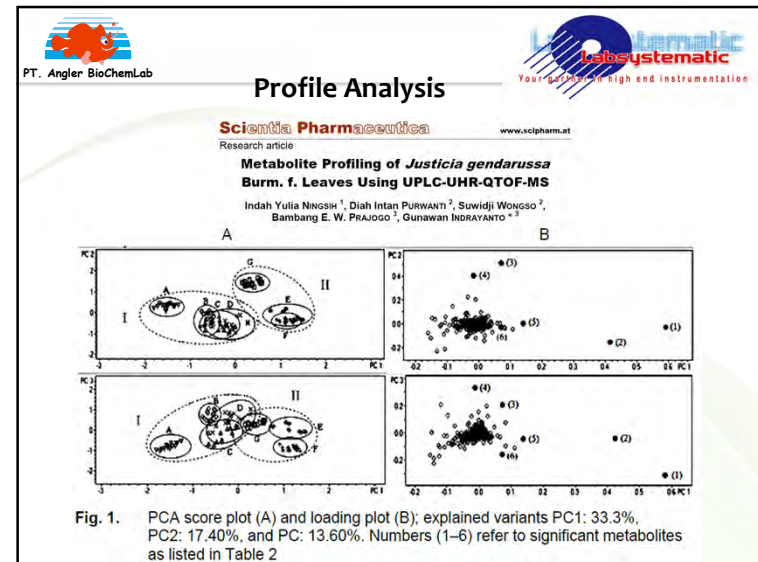
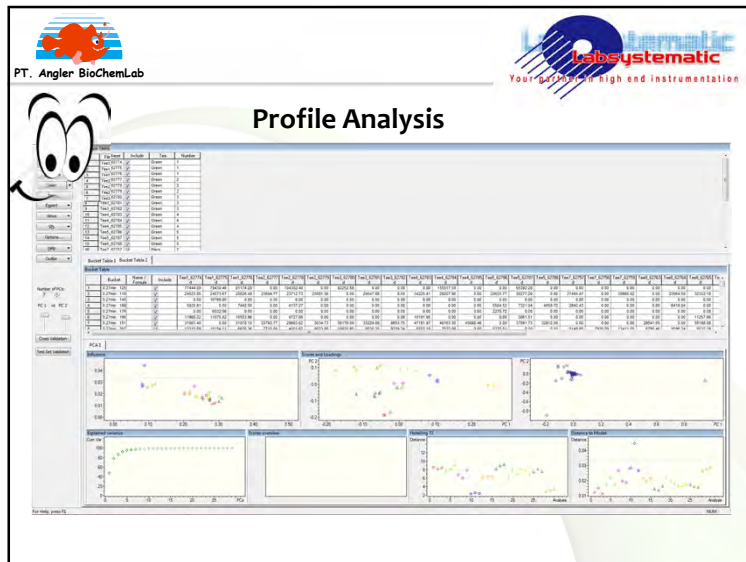
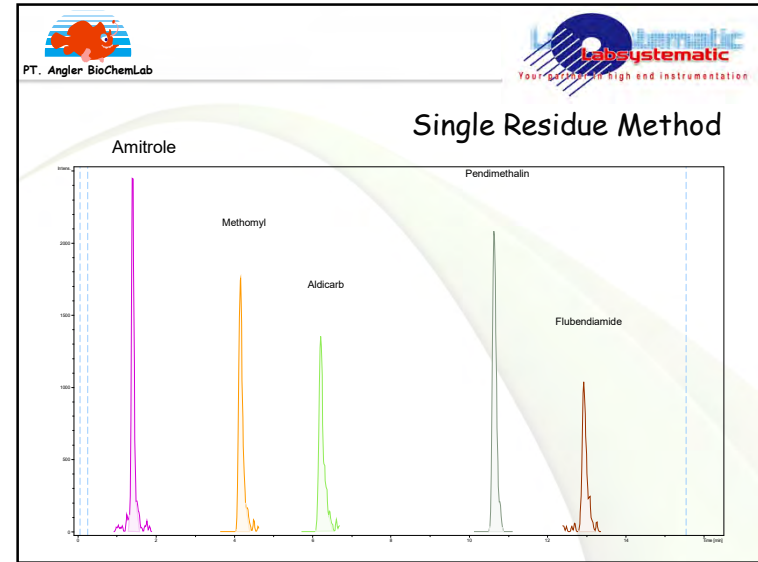
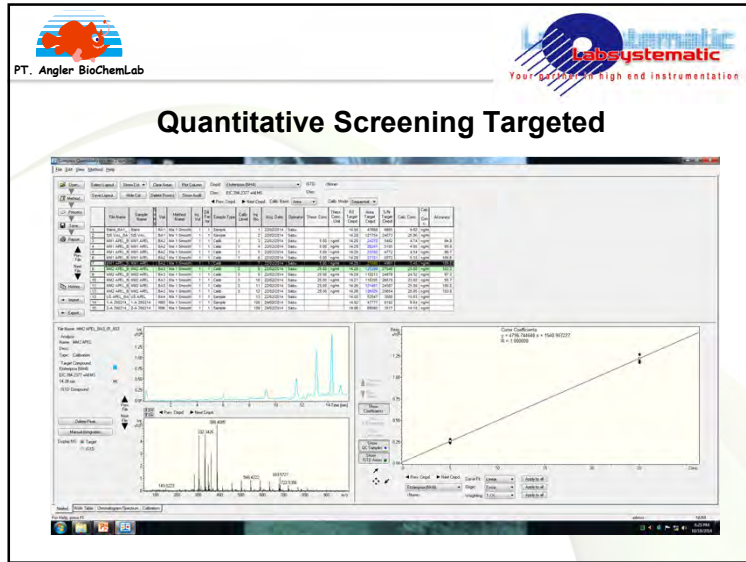
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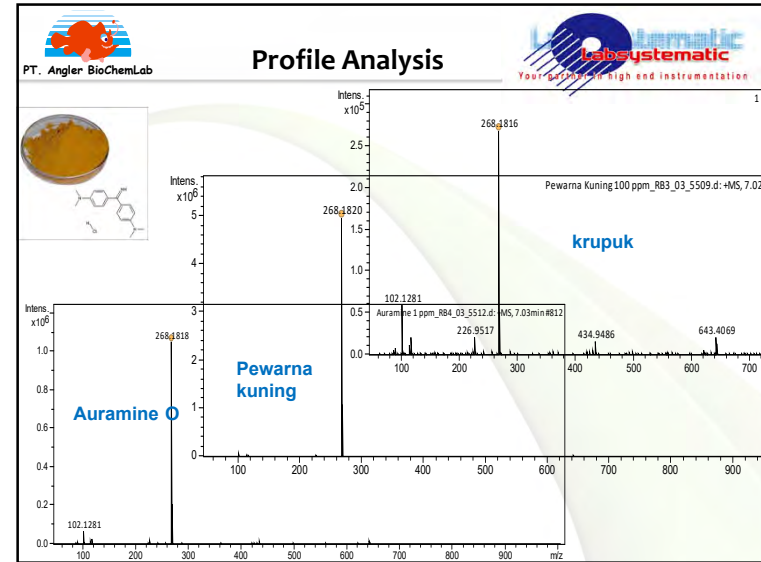
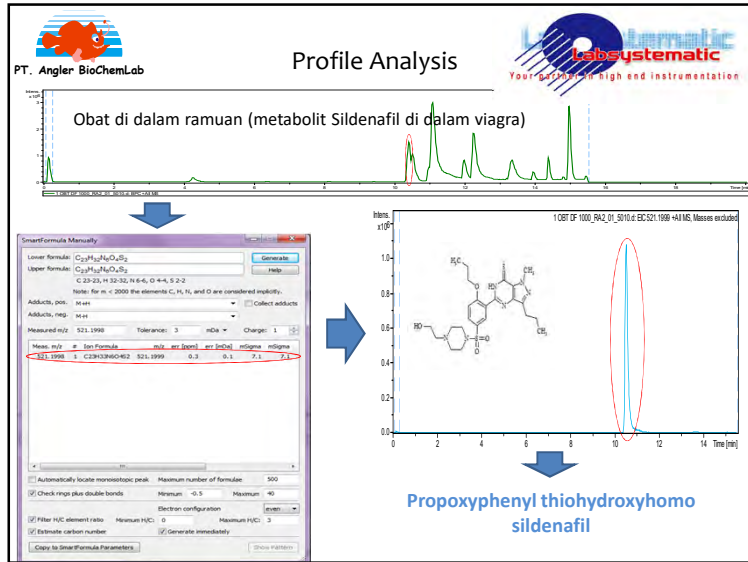
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Quantitative Screening Targeted



Hasil kromatogram dari Analisis Target pada konsentrasi 25 µg/kg





Contact Us
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nutraceutical
 food water marine product
 cosmetics pharmaceutical feed
 environmental soil

Committed to Global Standard

Jl. Sawo No. 17 -19, Bringin, Sambikerep
 Surabaya 60218, Indonesia
 Telp : 031 - 7456111
 Fax : 031 - 99161116
 Email :
marketing@anglerlab.co.id
info@anglerlab.co.id

Member of ALPI (Indonesian Food Laboratory Association)
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