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High-Throughput Screening for Bioactive Natural Compounds from Plant Extracts by HPTLC Hyphenations

ÁGNES M. MÓRICZ¹; DÁNIEL KRÜZSELYI¹; MARYAM JAMSHIDI-AIDJI²;
PÉTER G. OTT¹; GERTRUD E. MORLOCK²

¹ Plant Protection Institute, Centre for Agricultural Research, Herman O. Str. 15, H-1022 Budapest, Hungary

² Interdisciplinary Research Center (iFZ), and Institute of Nutritional Science, Chair of Food Science, Justus Liebig University Giessen, Heinrich-Buff-Ring 26-32, 35392 Giessen, Germany

Correspondence: moricz.agnes@agrar.mta.hu

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1. Introduction

In the last decade, effect-directed analysis (EDA) gave new impetus for the discovery of new potential drug compounds from natural sources. High-performance thin-layer chromatography (HPTLC) was established as a high-throughput and reliable separation technique that is frequently utilized for screening of highly complex samples, such as crude plant extracts. HPTLC combined with biological and biochemical assays and high-resolution mass spectrometry (HRMS) followed by bioassay-guided isolation and NMR was demonstrated as a straightforward strategy for bioanalysis of natural products [1,2].

Diabetes and Alzheimer's disease (AD) represent two of the global health issues. Type 2 diabetic patients, the majority of the people with diabetes, suffer from the hyperglycemia. The salivary and pancreatic α -amylases and α - and β -glucosidases are involved in the degradation and digestion of poly- and oligosaccharides to glucose, hence, glucosidase and amylase inhibitors are of therapeutic interest in type 2 diabetes as well as overweight and obesity [3,4]. AD is associated with the loss of cholinergic neurons in the brain and decrease in the neurotransmitter acetylcholine. Acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) are crucially involved in the hydrolysis of the acetylcholine into choline. Hence, AChE-selective and dual AChE/BChE inhibitors represented the first generation of medicines for the decrease of the AD progression rate [5]. The fight against infectious diseases, a major global public hazard, is also a challenge due to the

increase in the emergence of the (multi)drug-resistant microorganisms [6].

In this study (HP)TLC combined with direct α - and β -glucosidase, α -amylase and antimicrobial assays will be demonstrated for the comparison of the bio-profiles (the enzyme inhibitory and antibacterial potentials) of invasive goldenrod species.

2. Results

Solidago canadensis (Canadian goldenrod) root extract was introduced to the HPTLC-enzyme inhibitory assays, including the α -glucosidase, β -glucosidase, α -amylase, acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) assays. Two compound zones, detected as black zones by HPTLC after derivatization with the vanillin sulphuric acid, were active in all enzyme inhibitory tests (Figures 1 and 2). HPTLC-HESI-HRMS analysis of these compounds provided the same molecular formula $C_{20}H_{28}O_3$ that was confirmed by HPTLC-DART-HRMS and SPME-GC-MS.

After preparative isolation, the compounds were identified by NMR as solidagenone and pre-solidagenone (its two isomers) [7].

3. Conclusions

High-throughput, fast, relatively cheap HPTLC-bioassays were successfully used for the screening of bioactive plant components. HPTLC-HRMS and NMR of the isolates ensured the identification of the active compounds that showed anti-hyperglycemic and anti-cholinesterase activity. Thus, they can be potential candidate for drug discovery in the future.

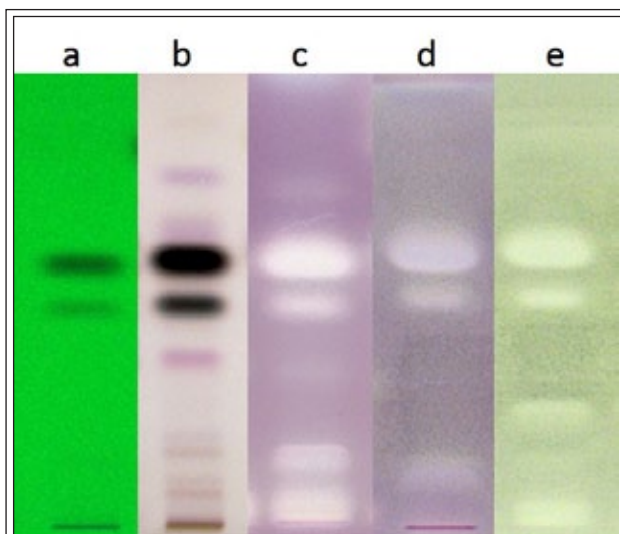


Figure 1 HPTLC chromatograms of bioactive components in the *Solidago canadensis* root extract at UV 254 nm (a) and white light after vanillin-sulphuric acid derivatization (b), as well as after α -glucosidase (c), β -glucosidase (d) and α -amylase (e) assays. Bright zones in the bioautograms indicate the bioactive compounds.

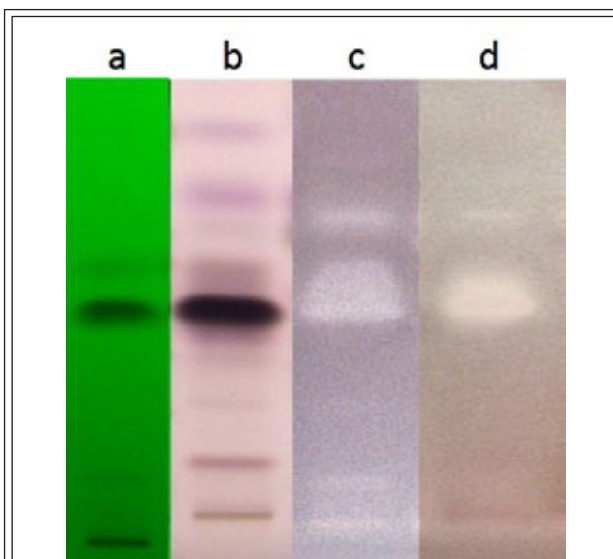


Figure 2 HPTLC chromatograms of bioactive components in the *Solidago canadensis* root extract at UV 254 nm (a) and white light after vanillin-sulphuric acid derivatization (b), as well as after AChE (c) and BChE (d) assays. Bright zones in the bioautograms indicate the bioactive compounds.

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