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SOPHORA JAPONICA L. GUL KURTAKLARIDAN AJRATIB OLINGAN RUTIN VA KVERSETINNI BIR VAQTDA ANIQLASHNING SPEKTROFOTOMETRIK USULINI ISHLAB CHIQISH

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Annotatsiya: *Sophora Japonica L.* gul kurtaklaridan ajratib olingan rutin va kversetinni bir vaqtda aniqlashning oddiy, tez, aniq va iqtisodiy spektrofotometrik usuli ishlab chiqilgan. Rutin va kversetin mos ravishda 258 va 374 nm da maksimal absorbsiyani ko'rsatganligi sababli, mos ravishda rutin va kversetinni aniqlash uchun ushbu to'liq uzunliklarida absorbsiya o'lchandi. Rutin va kversetin 1-10 mkg/ml kontsentratsiya oralig'ida Beer-Lambert qonuniga bo'ysunadi. Usul rutin va kversetinni bir vaqtning o'zida muntazam ravishda aniqlash uchun qabul qilinishi mumkin.

Kalit so'zlar: *Sophora Japonica L.*, Rutin, Kversetin, UB, IQ spektroskopiya, Kalibrlash egrisi, ekstraktsiya.

DEVELOPMENT OF A SPECTROPHOTOMETRIC METHOD FOR SIMULTANEOUS DETERMINATION OF RUTIN AND QUERCETIN ISOLATED FROM *SOPHORA JAPONICA L.* FLOWER BUDS

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Abstract: A simple, fast, accurate, and economical spectrophotometric method has been developed for the simultaneous determination of rutin and quercetin isolated from *Sophora Japonica L.* flower buds. Since rutin and quercetin show maximum absorbance at 258 nm and 374 nm, respectively, absorbance measurements were taken at these wavelengths for their determination. Rutin and quercetin obey the Beer-Lambert law within the concentration range of 1-10 µg/ml. This method can be adopted for the routine simultaneous determination of rutin and quercetin.

Keywords: *Sophora Japonica L.*, Rutin, Quercetin, UV, IR spectroscopy, Calibration curve, Extraction.

РАЗРАБОТКА СПЕКТРОФОТОМЕТРИЧЕСКОГО МЕТОДА ОДНОВРЕМЕННОГО ОПРЕДЕЛЕНИЯ РУТИНА И КВЕРЦЕТИНА, ВЫДЕЛЕННЫХ ИЗ БУТОНОВ *Sophora Japonica L.*

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Аннотация: Разработан простой, быстрый, точный и экономичный спектрофотометрический метод одновременного определения рутина и кверцетина, выделенных из бутонов *Sophora Japonica* L. Поскольку рутин и кверцетин показывают максимальное поглощение при 258 и 374 нм соответственно, измерения поглощения проводились на этих длинах волн для определения рутина и кверцетина соответственно. Рутин и кверцетин подчиняются закону Бера-Ламберта в диапазоне концентраций 1-10 мкг/мл. Метод может быть применен для одновременного рутинного определения рутина и кверцетина.

Ключевые слова: *Sophora Japonica* L., Рутин, Кверцетин, УФ, ИК-спектроскопия, Калибровочная кривая, экстракция.

KIRISH

Flavonoidlar o'simliklar dunyosida keng tarqalgan birikmalar bo'lib, ular antioksidant, yallig'lanishga qarshi, *saratonga qarshi faollik, neyroprotektiv ta'sirga ega* [1-3]. Flavonoidlarning eng keng tarqalgan vakillaridan rutin va kversetin o'zining ko'plab biologik ta'sirlari tufayli keng qo'llaniladi.

Hozirgi vaqtda rutinni o'simlikning yer ustki qismidan va yapon saforasining (*Sophora japonica* L.) gul g'unchasidan olinadi [4]. Olingan rutinni mineral kislotalar yordamida gidroliz qilish orqali esa kversetin olinadi [5; 6].

Adabiyotlar tahlili shuni ko'rsatadiki, Rutin va kversetinni sifat va miqdoriy aniqlashning YuQX, UB-spektroskopiya, YuSSX kabi usullari tavsiya etilgan [7-9].

Shu boisdan rutin va kversetinning yangi manbaalarini izlab topish va ularni bir vaqtning o'zida aniqlashning tezkor usullarini ishlab chiqish muhim vazifa hisoblanadi.

MATERIALLAR VA METODLAR

Reaktivlar: Kversetin ($\geq 97.0\%$; 302.24 g/mol) va rutin ($\geq 95.0\%$; 610.52 g/mol) Sigma-Aldrich® Co. (Sent-Luis, MO, AQSh) kompaniyasidan xarid qilingan, 96% etanol ishlatildi.

UB va IQ spektroskopiya sharoitlari. Kompyuter dasturi UV Probe 2.0 bilan ulangan (spektral kengligi 2 nm, to'lqin uzunligi aniqligi 0.5 nm) ikki nurli UV-spektrofotometr (Shimadzu, UV-1900i, Yaponiya) va 1 sm.li juftlashgan kvarts kyuvetalar va raqamli tarozi (Mettler Toledo, AB265-S/FACT, Shveysariya) taklif etilayotgan analitik usulni ishlab chiqish va validatsiya qilish uchun ishlatildi.

Ekstraktlarni tayyorlash. Rutin yapon soforasi gullaridan suvli ekstraktsiya va etil spirtida qayta kristallash orqali ajratib olindi. Olingan rutin mineral kislotalar yordamida gidrolizlanib, kversetin hosil qilindi [10-11].

Standart eritmalarini tayyorlash va kalibrlash egri chizig'ini tuzish. Rutin va kversetinning standart eritmaları har bir moddadan 10 mg olinib etanolga eritildi va hajmi shu erituvchi bilan 100 ml li o'lchov kolbasida 100 mkg/ml konsentratsiyaga yetkazildi. 10 mkg/ml li ishchi standart eritmasi $\lambda_{\max}$ UV spektrofotometrda (600–200 nm) aniqlandi. Beer-Lambert qonunini va mos ravishda rutin va kversetin uchun regressiya tenglamalarini o'rganish uchun konsentratsiya va absorbsiyaga bog'liq kalibrlash egri chiziqlari tuzildi.

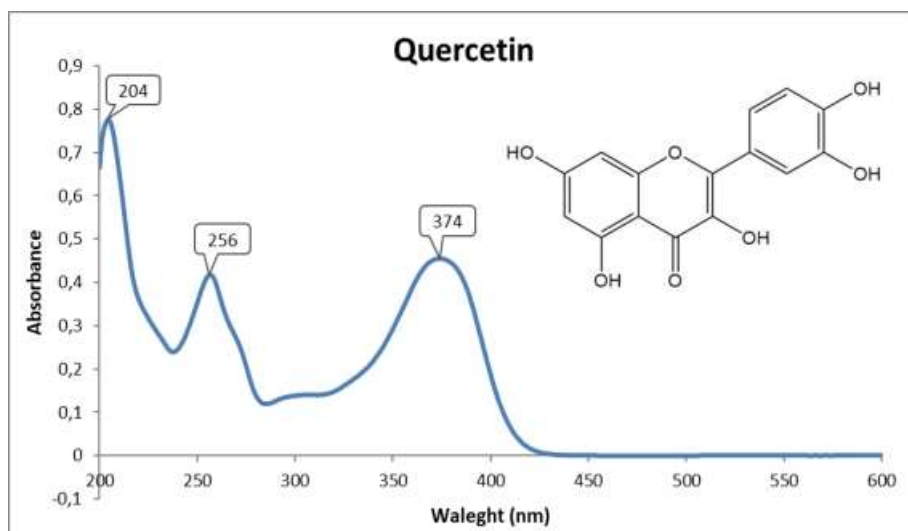
NATIJALAR VA MUHOKAMA

Rutin va kversetin uchun chiziqli diapazon tegishli tanlangan to'lqin uzunliklarida 2-20 mkg/ml ni tashkil etadi. 257 nm da rutin uchun va 372 nm da kversetin uchun korrelyatsiya koeffitsienti mos

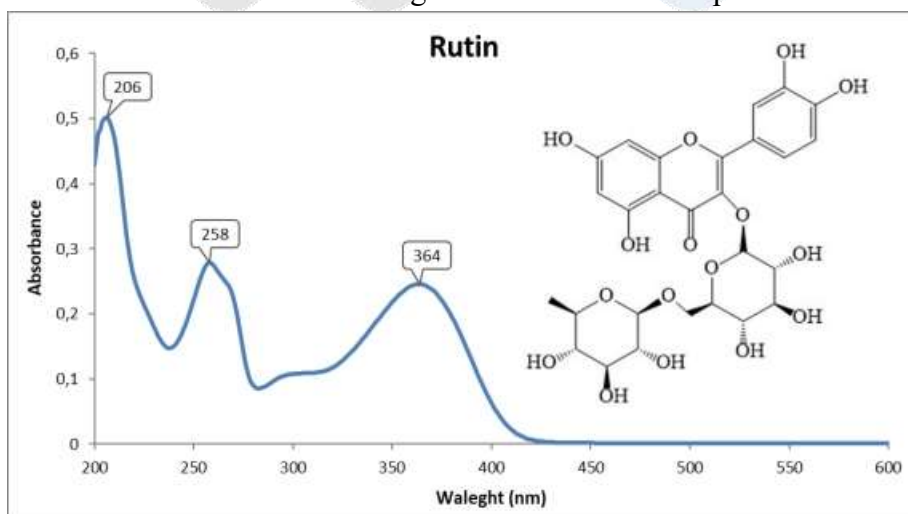
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ravishda 0,997 va 0,992 ni tashkil etadi. Ikkala modda ham o'zlarining tegishli to'liq uzunliklarida yaxshi regressiya qiymatlarini ko'rsatadi va tiklash tadqiqotining natijalari shuni ko'rsatadiki, eritmadagi dori konsentratsiyasidagi har qanday kichik o'zgarish taklif etilgan usullar yordamida aniq aniqlanishi mumkin.

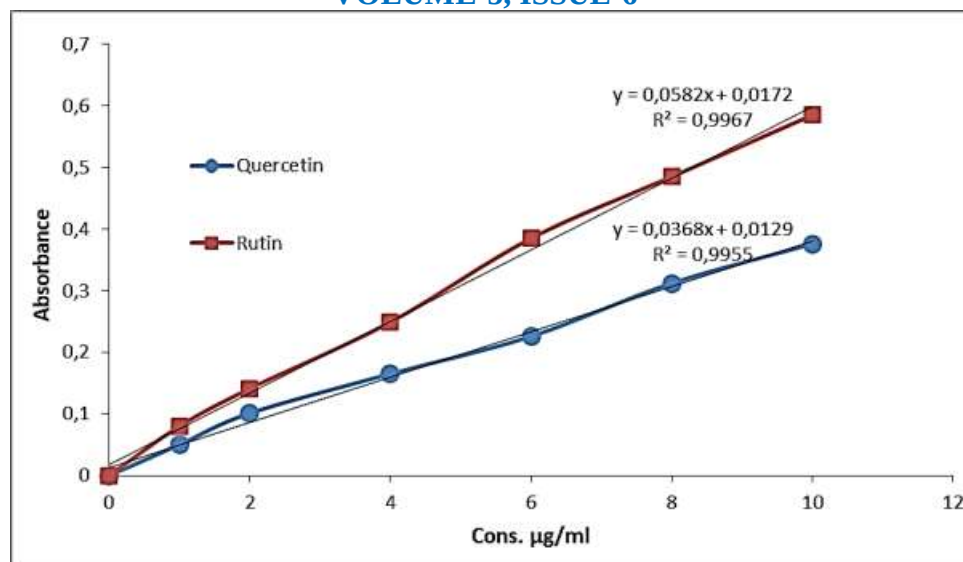


1-rasm. Kversetinning etanolli eritmasi UB spektri.



2-rasm. Rutinning etanolli eritmasi UB spektri.

Usulning aniqligini aniqlash uchun rutin va kversetinning olti xil konsentratsiyasining (1, 2, 4, 6, 8, 10 $\mu\text{g/ml}$) optik qiymatlari uch marta qayta o'lchandi. Olingan natijalarga asoslanib, rutin va kversetinning kalibrlash egri chizig'i va kalibrlash tenglamasi tuzildi (3-rasm).

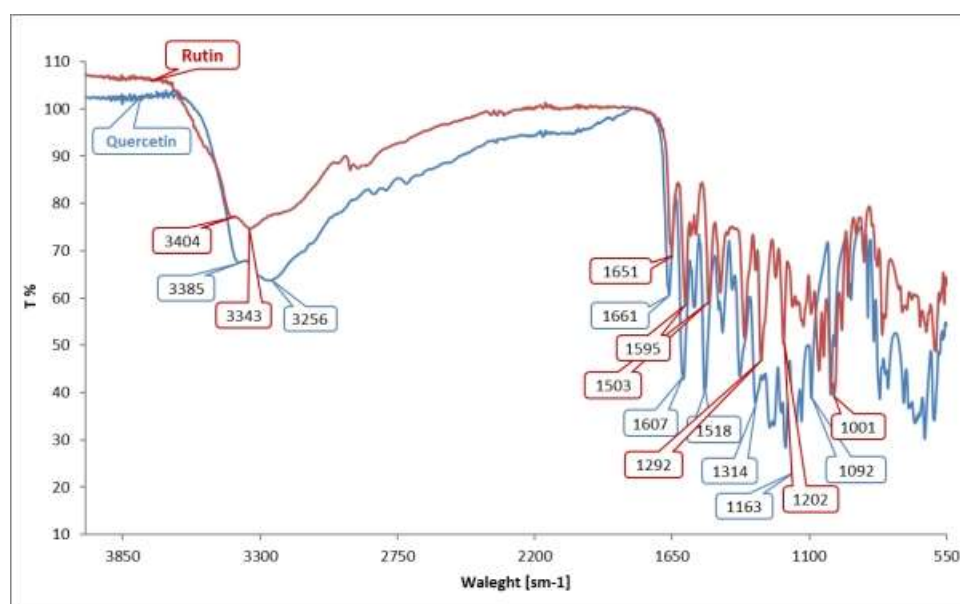


3-rasm. Kversetin va Rutin uchun kalibrlash grafigi

Har bir modda uchun standart eritmalaridan mos ravishda tayyorlangan suyultmalar ishlab chiqilgan usullar asosida tahlil qilindi. Beer-Lambert qonuni asosidagi konsentratsiya diapazoni Rutin uchun 1–10 µg/ml va kversetin uchun ham 1–10 µg/ml deb aniqlandi. Ushbu usul bo'yicha chiziqlilik (linearlik) ma'lumotlari 1-jadvalda keltirilgan.

1-jadval. Validatsiya parametrlarining natijalari

Parameter	Rutin	Kversetin
To'lqin uzunligi (nm)	258	374
Chiziqlilik diapazoni (µg/ml)	1-10 µg/ml	1-10 µg/ml
Chiziqli tenglama	$y = 0,0582x + 0,0172$	$y = 0,0368x + 0,0129$
Korrelatsiya (R^2)	0,9967	0,9955
Qiyalik (b)	0,0582	0,0368
Kesishma (a)	0,0172	0,0129



4-rasm. Kversetin va rutinining IQ spektrlari

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Jadval 1. Kversetin va rutinning IR spektrlari (to'liq soni sm^{-1} da)

Birikma	$\nu(\text{C}=\text{O})$	$\nu(\text{C}=\text{C})$	$\nu(\text{O}-\text{H})$	$\nu(\text{C}-\text{O}-\text{H})$	$\nu(\text{C}-\text{O}-\text{C})$
Kversetin	1661	1607, 1518	3385-3256	1314	1163, 1092
Rutin	1651	1595, 1503	3404-3343	1392	1202, 1084

XULOSA

Tavsiya etilayotgan spektrofotometrik usul oddiy, tezkor, aniq, to'g'ri va iqtisodiy bo'lib, chiziqlilik, aniqlik, to'g'rilik, spetsifiklik va takrorlanuvchanlik bo'yicha validatsiyadan o'tgan. Ushbu usulni birikmalar tarkibidagi rutin va kversetinni bir vaqtning o'zida aniqlash uchun muvaffaqiyatli qo'llash mumkin.

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