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Precision Medicine by Combining Diagnosis with Prophylaxis Towards "Prophynostics": Controlled Delivery of Nicotine using a Device Platform for Treating Epilepsy

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Precision medicines are next-generation medicines that apply logistics to improve the efficacy of therapy, reduce safety issues, and reduce patient non-compliance. In this line, we propose a new concept of combining diagnostics with prophylaxis and token the idea of "prophynostics." Detection followed by prophylactic therapy can prevent disease occurrence. That is particularly important in the case of situations like preventing COVID-19 viral spreading and seizure prevention in epilepsy. However, efforts to achieve this goal is somewhat elusive. This work reports a device platform for detection, followed by the delivery of nicotine through two different routes. Modeling of the detection of a suitable clinical situation is done by choosing epileptic seizure detection. Subsequently, we designed and developed a vibration dependent detection device for detecting seizure type vibrations. For attaining rapid loading dose in patients, delivery through the sublingual route is planned. For that, appropriate nicotine, aspirin, paracetamol sublingual patch is designed and developed. To achieve prolonged delivery of a sustained dose of nicotine in patients, transdermal delivery is planned. For that, suitable nicotine, aspirin, and paracetamol transdermal patch are designed and developed. The platform possibilities of the delivery systems are demonstrated using drugs from various BCS classification system. The drug release properties of the optimized patches are fitted with different models such as zero order, first order, Higuchi, and Koresmayer-Peppas and optimized the release profile as per requirements. This combined system can overcome issues like addiction possibilities of nicotine while exploring it for prophylaxis. Nicotine proposed to have anti-COVID and antiepileptic properties in addition to the anti-inflammatory properties of NSAID's; for such purpose, this type of device can be developed and used.

Introduction

Precision medicine is aimed to improve the efficiency of therapy by reducing the complications and safety issues while enhancing the efficacy of treatment [1]. Many strategies have recently emerged as next-generation technologies such as theranostics [2], feed-back regulated drug delivery systems [3], and various genetic platforms [4].

Here, the detection of disease condition followed by therapy is a possibility for theranostics [5], which is an example of precision medicine [6]. On the other hand, detection followed by prophylaxis is also possible, which can lead to a new area of "prophynostics" [7-9]. From that perspective, this work is the first report where detection is combined with prophylaxis.

COVID-19 viral infections have a high mortality rate in the case of infected patients, particularly one with comorbid conditions [10]. It's a fast-spreading pandemic since there are no particular medicaments available for the therapy of COVID-19 [11], prophylactic strategies are gaining importance [12] where various drugs are to be administered for rapid onset of action followed by prolonged delivery [13].

In the case of epilepsy, nocturnal seizure in neonates and infants are trending to be life-threatening by 57-137 per 1000 live birth per year [14]. In this case, seizure attacks majorly occur during Non-Rapid Eye Movement (NREM) sleep [15]. Seizure occurring during NREM is fatal [15]. This syndrome [16] is due to missense mutation in the alpha subunit of nicotinic acetylcholine receptor

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[17,18] and is widely known as Autosomal Dominant Nocturnal Frontal Lobe Epilepsy (ADNFLE) [19,20]. During sleep, since both NREM and REM (Rapid eye movement) patterns occur repeatedly, the seizure is painful to detect [21]. In this condition, nocturnal management of seizure is vital by rapid onset of action followed by controlled delivery throughout the night.

Nicotine is the ideal candidate drug that can be explored as a candidate drug for precision medicine as it causes addiction [22]. It is now proposed as a drug to treat COVID-19 infection [23] and epilepsy [24]. The severe acute respiratory syndrome coronavirus has ACE-2 (Angiotensin Converting Enzyme-2) as their host receptor present in the airways, heart, bladder, and olfactory areas of the body. The enzyme is essential in Renin Angiotensin Aldosterone System (RAAS) for regulating blood pressure and fluid balance in the body. Nicotine acts as a nicotinic acetylcholine receptor agonist that is capable of up-regulating ACE-1, Angiotensin (ANG)-II, and ANG II (type 1) receptor axis. It also simultaneously down-regulates ACE-2, ANG-(1–7), and Mas receptor axis. By the administration of nicotine, the down-regulation of ACE-2 leads to fewer host receptors for COVID binding and can be useful as a preventive tool in preventing the pandemic as well [25-27]. Apart from nicotine NSAIDs such as aspirin and paracetamol are useful for reducing the inflammation in COVID-19 [28].

On the other hand, nicotine can modulate mood shifts by activating the areas involving mood changes and feelings of pleasure. The excitement of these nicotinic acetyl cholinergic receptors (nAChRs) leads to the release of other neurotransmitters, such as dopamine (also known as pleasure hormone). It alters the mood shift that is beneficial in the suppression of epilepsy. The seizure attack occurring during sleep in infants and neonates are fatal. The drug can be provided in treating the same by altering the neurotransmission mechanism [29].

From a personalized medicine perspective, nicotine needs to be delivered with care in a dose-controlled manner. For broader application to a large sector of patients, with minimum clinical motivation, nicotine needs to be delivered non-invasively through suitable routes. Also, if required, the therapy needs to be withdrawn.

Detection systems explore various physical and chemical platforms [30]; wherein, any of the physical or chemical markers are detected nullifying the error contribution [31]. For example, in the case of epilepsy, the current detection systems are based on (a) nerve impulse detection [32], (b) exchange of impulses [33] and (c) to study the emulating changes in the brain as well as in the systems as a whole [34]. Vibration-based seizure detection are particularly useful for the neonatal patients [34, 35]. The detection device prototypes of various configurations are being developed as per patient requirements [36]. Cradle fitting detection devices are developed for neonates [37]. For seizure detection, seizure type vibrations are specifically detected [38]. Similarly, for the detection of COVID-19 various RT- PCR detection methods are available [39].

For the delivery of suitable nicotine route of administration, meeting set requirements are to be identified, and suitable technologies need to be explored. Controlling the rate of drug delivery is highly mandatory because of its carcinogenic [40] as well as addiction [41] history. For the rapid onset of action, the sublingual route is most preferred for the delivery of a given loading dose [42]. For that, sublingual patches are most suitable [43]. Prolonged-release of the sustained dose is achieved through the transdermal route using a transdermal patch [44]. By adjusting the disintegration and dissolution of the patch the required release rates are achieved [45]. The patches are characterized for physical appearance, drug content, and drug release properties. The versatility of the drug release platform is analyzed by performing drug release using various drugs from the BCS classification system.

If there is a system to detect and after detection, if the nicotine can be delivered using one or multiple systems for rapid onset and the prolonged effect it can address some of the cardinal patient issues, such as treatment could be possible with less motivation in the case of nocturnal seizure attacks in epileptic patients and probably prevention of COVID-19 spreading. A prototype is designed and developed here that involves the development of a combination system to detect the nocturnal seizure followed by therapy using nicotine.

Subsequently, following systems are developed, including (a) a cradle fitting model of seizure type vibration detector [35], (b) a fast-dissolving sublingual patch that delivers the entire drug within 5min of administration for providing rapid onset and reproducibility, through the oral cavity [33] and (c) a transdermal patch for prolonged drug delivery [44] up to 6hours to prevent the seizure episodes during sleep in epileptic patients. The designed and developed systems are eventually characterized by its consistency and robust nature.

Materials and Methods

For detection device prototype

Voltage regulator (from Core Embedded Solutions, Haryana), Vibration sensor (Adraxx electronics, New Delhi), Battery, Buzzer, LED, Switch, IC (3 types from Findx Pro, Bengaluru), Transistor BC 547 (Thomson electronics, Kochi), Resistor (Mascot Computers, Mumbai), Capacitor (Sumeru Labs, Bangalore), Battery Connector (Plan My Study, Nagpur), Printed circuit board, AT-mega 328 (Microchip Technology Pvt. Ltd, Pune), 16 MHz microcrystal (Orbit electronics, Mumbai), IC Base (Microchip Technology Pvt. Ltd, Pune), Copper Clad Laminate (Epoxy house, Delhi), Ferric chloride powder (Labdhi chemicals, Mumbai), 10k 0.25 CFR (Mascot Computers, Mumbai), Cardboard, Gold paper, and plastic box is purchased from the local electronic shop.

For patches

Hydroxy Propyl Methyl Cellulose (Loba chemicals Pvt ltd. Mumbai), Poly Ethylene Glycol(4000) (Loba chemicals Pvt ltd. Mumbai), Propylene Glycol (Nice), Sodium Starch Glycolate (Yarrow chem products, Mumbai), Methanol (Spectrum, Kochi), Ethanol (Spectrum, Kochi), Dichloromethane (Spectrum, Kochi), Nicotine (Hi media laboratories Pvt.Ltd, Mumbai), Aspirin (Spectrum, Kochi), Paracetamol (Research lab fine chem industries, Mumbai), Hydrochloric acid (Spectrum, Kochi), Sodium Hydroxide pellets (Spectrum, Kochi) and potassium dihydrogen orthophosphate (Spectrum, Kochi).

Design and development of device prototype for seizure detection

The cradle fit model was developed and validated for producing alarm only in the seizure type vibrations for which it was exposed to variant types of movements like shaking or jerky, rotation, and oscillations. The sensitivity of the device in terms of percentage accuracy in making alarms during any unwanted motions was studied systematically. The prototype was a digital circuit, which uses two IC chips (555IC and a 74193 counter IC). In this circuit, the vibration sensor produces pulses during the vibration. This analog signal was converted to a digital pulse using the 555IC as a mono-stable configuration. The digital pulses were fed to the counter

IC, where it increments the count on each digital pulse. When the count reaches 16, it produces a single pulse output. This output was fed to the second 555 in the mono-stable configuration. So the second 555 triggers and produces a long pulse output. This output pulse triggers the LED and buzzer.

Preparation of sublingual patch [46,47]

Sublingual patches were prepared as per the solvent casting method. Hydrophilic polymer (HPMC) (2 gm), PEG-400 (1g), and a super disintegrating agent (sodium starch glycolate) (2.5g) were taken respectively and mixed well in the solvent combination (Dichloromethane and Ethanol each 7 ml). The mixture was spread evenly onto the glass slide pre-coated with glycerine and kept to dry under 60ºC in a hot air oven. The patches after being optimized were loaded with different drugs during mixing (Aspirin, Paracetamol, and Nicotine) and solvent cast to study various parameters.

Preparation of transdermal patch [48]

Hydroxypropyl methylcellulose (1 g), PEG-400 (0.25 mg), Propylene glycol (0.4 ml) were taken respectively and dissolved in methanol (15 ml). The obtained transparent mixture was further poured onto the previously glycerine coated Petri dish and allowed to dry at 75ºC in a hot air oven. The patches loaded with different drugs (Aspirin, Paracetamol, and Nicotine) were taken further for establishing its properties.

Evaluation of the vibration detection prototype

The developed prototype was subjected to oscillatory, rotatory, and shaking or jerk-like movements seen similarly in a cradle. The developed prototype was carefully fit into the cradle, and with the help of 7 random volunteers, it was oscillated to note the time at which it alarms. The same was repeated with another set of 7 individuals to rotate the cradle clockwise direction to check the maximum rotation at which the device develops alarm. The final evaluation of the device for shaking or jerk-like movements mimicking the epileptic seizure attack was performed by random individuals to check the alarm production threshold. The above said, an evaluation was performed thrice with the change in individuals to attain robustness and reproducibility in the developed device prototype.

Characterization of patches

Organoleptic properties such as odor, nature, color, the appearance of the sublingual and transdermal patches were observed and recorded. Different drug-loaded patches were weighed using an electronic balance, and weight variation [49] were calculated. Patches of desired measurement 1 x 1 cm was cut and folded several times until it breaks into two. The number of successful folds was recorded and noted down to calculate folding endurance [50]. Patches of the desired length were cut and kept in a desiccator containing calcium chloride for 3days. Weight was taken and the percentage of moisture loss (PML) [51] calculated. Drug loaded patches (1x1cm) were uniformly cut and placed onto desiccator containing a saturated solution of aluminum chloride for 79.5% humidity. After 3days, the patches were removed and weighed to calculate percentage of moisture absorbed (PMA) [46]. Patches (1x1cm) were cut and weighed accurately. Patches were immersed onto phosphate buffer solution (PBS) (pH 6.8 and 7.4) in a Petri dish. After 5 mins, the patches were removed and weighed, and the swelling index (SI) [51] was calculated using the below formula.

SI = [(final weight - initial weight)/initial weight] x 100

Drug content uniformity [52] was calculated as follows. Respective patches were uniformly cut into 1x1 cm and were dissolved in 100ml phosphate buffer (pH 6.8 and 7.4). The sample was further analyzed at l_{max} 248 nm (paracetamol), 254 nm (aspirin), 259 nm (nicotine).

***In vitro* release study of sublingual patches [52]**

Patches were cut uniformly into 1x1 cm and were released into a 200ml buffer solution (pH 6.8). The study was performed using the dissolution apparatus (type II) USP conserved at 37+0.5ºC with75 rpm. At regular intervals of 1 min, 2 min, 3 min, 4 min, and 5 min, 5ml of the solution was withdrawn and replaced with a buffer solution (pH 6.8). Absorbance was noted for specific drugs at respective wavelengths at l_{max} Aspirin (248nm), Paracetamol (254nm), and Nicotine (259nm). A graph was plotted between % drug release and time.

***In vitro* release study of transdermal patches [53]**

The patches were cut into 1x1 cm and released into buffer solution of pH 7.4. The study was performed using a magnetic stirrer using a cellophane membrane attached to an open-end tube. The receptor compartment was continuously agitated magnetically, with the temperature at 32 ± 0.5ºC. The samples withdrawn, are analyzed for its content by UV Spectrophotometry for paracetamol [248nm], aspirin [254nm], and nicotine [259nm].

***In vitro* release study of combination patches**

The prepared sublingual and transdermal nicotine patches were placed into two distinct open-end tubes held over a beaker containing 100ml buffer solution on a magnetic stirrer and maintained at room temperature, and successive volume of samples was collected at definite intervals to calculate the percentage release of the drug.

Kinetic modelling study [54-56]

The regression coefficient (R) denotes the linearity of the plots. The plots containing the highest linearity is chosen as the most appropriate model of choice.

A zero-order release is denoted by

$$Qt = Qo - Kot$$

where, Qo = initial amount of drug, Qt = the amount of drug release at time 't', Ko = rate constant. A graph of cumulative percentage of drug released vs. time provides a straight line with a slope equal to Ko.

First-order, denotes the reaction to be concentration-dependent. Thereby it can be equated as,

$$\ln Q = \ln Qo - K_1 t$$

where, K_1 is the release constant. A plot between log cumulative percentage drug retained and time would fetch the graph.

In Higuchi model, the pattern of drug release from the developed system can be predicted by,

$$Q = K_H . t^{1/2}$$

where K_H is Higuchi dissolution constant, 't' is the time in hours. The release data is plotted with a cumulative percentage drug release against the square root of time.

Korsmeyer-Peppas model explains the mechanism of drug release from the delivery system with a plot between the log cumulative percentage of drug release and log time. The 'n' was calculated through the slope obtained. Korsmeyer-Peppas equation as in

follows,

$M_t/M_a = Kt^n$

for, M_t/M_a = Fraction of solute released, 't' = Time, K = Kinetic constant, and 'n' is an exponent indicating the drug-releasing mechanism.

For every n <0.5, the mechanism followed is the diffusion of the drug from the reservoir. In case n lies between 0.5 and 1, the release follows an anomalous or non-fickian mechanism. A zero-order [case 2] release is found when n is equal to 1, and a non-fickian super case 2 release is depicted in cases with n greater than 1.

Statistical analysis

All the experiments were done with a minimum of three samples, wherever more samples are done, it is mentioned in the text - the data represented as the mean ± standard deviation.

Results and Discussion

Here we aim to develop a device prototype capable of alarming when a seizure occurs, along with a fast-dissolving sublingual patch for delivery of loading dose and a transdermal patch for delivery of the sustained dose of nicotine. The combined system contains these three parts: (i) device prototype, (ii) a sublingual patch, and (iii) a transdermal patch.

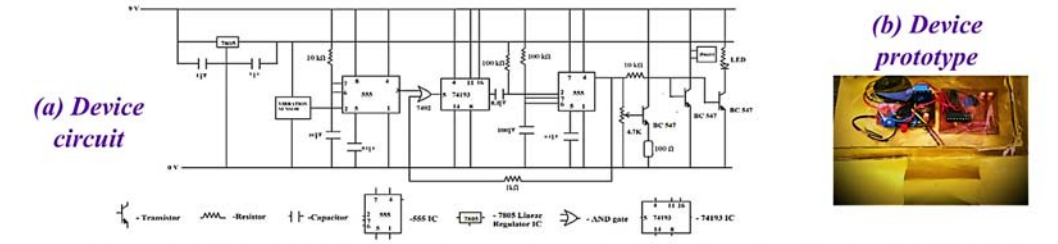
Device prototype for seizure detection

Using sensor, battery, switch, buzzer, and the alarming unit, a cradle fitting vibration-based seizure detection device prototype was developed as per the configuration in figure 1a as seen in figure 1b.

The vibration detection device is subjected to evaluation for different types of movements such as oscillation, rotation, and seizure type vibration. To check the response of the device to oscillations, the device is oscillated 400 times by seven different individuals that gave no alarm, as given in figure 1c. It shows that the device does not respond to oscillatory movements. Oscillatory movements are possible for the cradle, and the device needs to have the accuracy not to respond to such vibrations. Further, the device prototype is subjected to rotation type movements to find any response. As in figure 1d, the device, when rotated by another set of seven individuals, it required 30-39 rotations/min to produce any alarm. When the number of rotation increases beyond 20nos, it mimics vibratory movements, and rotatory movements are generally not possible in the case of cradle movements where generally oscillatory movements are possible. Given that, seizure type vibratory movements are set to analyze. The response of the device to seizure type vibrations are analyzed by subjecting the device to jerking movements by seven random individuals, and the results are shown in figure 1e. When analyzed for seizure type jerking movements, the device produces alarm with just 16-19 jerks, making the device suitable for identification of seizure type vibratory movements. After proving the accuracy of the detection device, the design and development of drug delivery patches are done. For that, first, for delivery of loading dose, a fast disintegrating sublingual patch is developed.

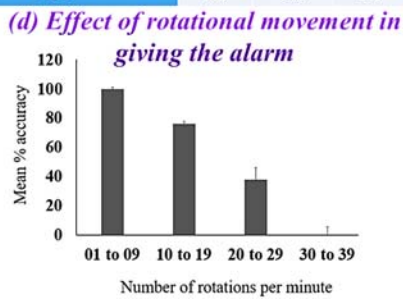
Sublingual patch

The formulation of the sublingual patch is developed by optimizing the composition of the patch as per the figure 2a as seen in figure 2b. For developing a sublingual patch with required properties, 18 formulations are developed (T1 to T18). The presence of glucose/lactose (added to improve solubility) led to a sticky non-transparent patch. The addition of Sodium starch glycolate (SSG) led to faster disintegration of a patch than usual. However, the concentration of SSG needed to be optimized for adjusting all the required properties of the patch. Here, HPMC acts as the binding



(c) Effect of oscillatory movements on giving the alarm (n=7) (I= random individuals)

Number of oscillations	Effect of oscillatory vibrations (Yes/No)							% Accuracy
	I-1	I-2	I-3	I-4	I-5	I-6	I-7	
489	No	No	No	No	No	No	No	100



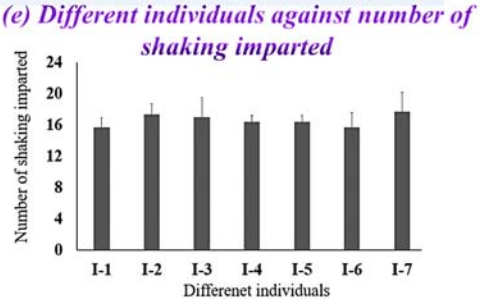


Figure 1: Vibration detection device configuration and cradle fitting prototype(a) Device circuit diagram, (b) device prototype, (c) Effect of oscillatory movements on giving the alarm (n=7)(I= random individuals), (d) Mean % accuracy vs. Number of rotations per minute graph(n=7), (e) Different individuals against the number of shaking imparted

(a) Composition of different formulations of sublingual patch towards its optimization

Trials	DCM (ml)	Ethanol (ml)	HPMC (g)	PEG-4000 (g)	Glucose (g)	Mg. St. (g)	SSG (g)	Methanol (ml)	Lactose (g)
T1	10	10	1	0.03	5	-	-	-	-
T2	10	10	1	0.5	2.5	-	-	-	-
T3	10	-	1	0.5	-	-		10	1.5
T4	10	10	3	0.5	-	-	0.15	-	-
T5	10	10	0.5	0.4	-	-	2.5	-	-
T6	5	5	0.5	0.4	-	-	2.5	-	-
T7	5	5	0.5	0.4	-	-	2.5	-	-
T8	3	3	0.5	0.4	-	-	2.5	-	-
T9	5	5	0.5	0.8	-	-	2.5	-	-
T10	5	5	0.8	0.8	-	-	2.5	-	-
T11	5	5	1	0.8	-	-	2.5	-	-
T12	Previous patch was coated with HPMC 0.3g and PEG 0.3g								
T13	5	5	0.8	1	-	-	2.5	-	-
T14	5	5	0.8	1	-	-	3	-	-
T15	5	5	0.8	1	-	0.5	2.5	-	-
T16	7	7	1	1	-	0.5	2.5	-	-
T17	7	7	0.8	1.2	-	0.5	2.5	-	-
T18	7	7	2	1	-	-	2.5	-	-

Abb:DCM: Dichloro methane, HPMC: Hydroxy methyl cellulose, PEG: Poly-Ethylene Glycol, Mg. St: Magnesium stearate, SSG: Sodium Starch Glycolate

(b) Image of the optimized sublingual patch

(c) Characterization of sublingual patches (n=3)

Evaluation of patches	Nicotine	Aspirin	Paracetamol
Folding Endurance	251.34 ± 1.24	251.67 ± 0.94	254.34 ± 0.5
Weight Variation	12.26 ± 0.29 mg	12.51 ± 0.31 mg	11.91 ± 0.12 mg
Percentage Moisture Loss	2.2% ± 0.02	1.98% ± 0.01	1.03% ± 0.03
Percentage Moisture Absorption	1.98% ± 0.01	2.02% ± 0.01	2.04% ± 0.01
Swelling Index	62.14% ± 0.03	68.15% ± 0.03	62.33% ± 0.07
Drug Content	0.03 ± 0.01 µg	0.04 ± 0.01 mg	0.02 ± 0.01 mg

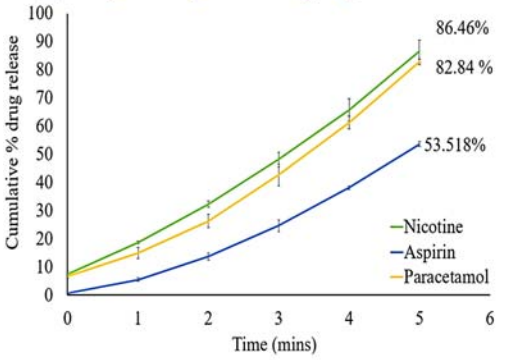
Figure 2: Development of sublingual patch, (a) Composition of different formulations of patch towards its optimization, (b) Image of the optimized patch, (c) Characterization of nicotine, aspirin and paracetamol patches respectively (n=3)

polymer when the amount is decreased, it creates a partially flexible patch that breaks after 10-15 folds. At higher concentrations, it forms a hard patch that breaks at the first fold. The amount of plasticizer is optimized with respect to the concentration of polymer to support the flexibility of the patch without affecting the binding property. The T18 formulation met all the required properties for a fast disintegrating sublingual patch. The picture of the optimized patch is given in figure 2b.

After optimizing the sublingual patch, it is evaluated for its physical properties. The physical properties of the patch loaded with different drugs such as nicotine, aspirin, and paracetamol is given in figure 2c. It shows that the patch irrespective of the loaded drug has required physical properties.

Subsequently, the patch is evaluated for its drug release properties. Figure 3 is the drug release profile showing the release of a maximum of drug 86.46%, 53.51 %, and 82.84 % within 5 mins for nicotine, aspirin, and paracetamol, respectively, showing the reproducibility of the product in attaining release of the required loading dose.

(a) Drug release from sublingual patches



(b) Regression coefficient of sublingual patches

Drug loaded patches	R^2				n
	Zero order	First order	Higuchi model	Korsmeyer-Peppas model	
Nicotine	0.95	0.79	0.76	0.99	1.59
Aspirin	0.94	0.76	0.73	0.99	2.5
Paracetamol	0.96	0.82	0.8	0.98	1.74

(c) Kinetic modelling of drug release from sublingual patches

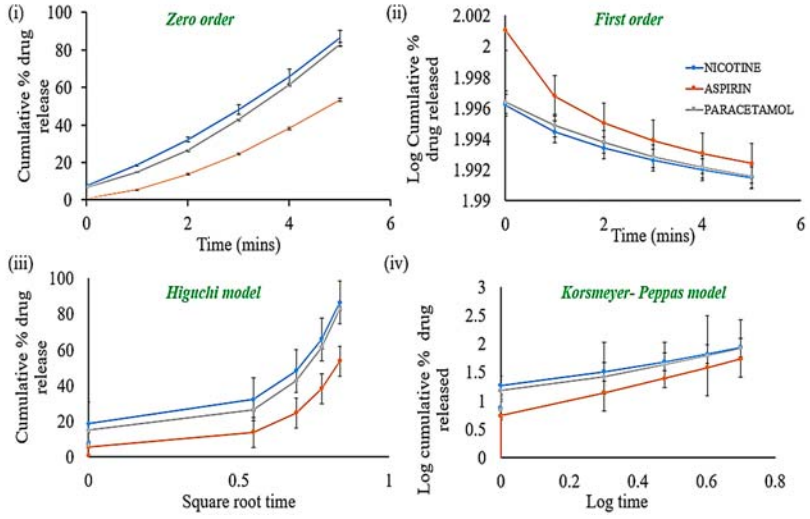
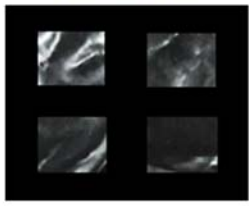


Figure 3: Drug release properties of the sublingual patch (a) Cumulative % drug release vs. Time graph of nicotine, aspirin and paracetamol sublingual patch respectively, (b) Regression coefficient of nicotine, aspirin, and paracetamol sublingual patches, (c) Graphs representing kinetic models of nicotine, aspirin, and paracetamol sublingual patches were; (i) Zero-order, (ii) First order, (iii) Higuchi model and (iv) Korsmeyer- Peppas model respectively

(a) Composition of different formulations of transdermal patch towards its optimization

Excipients	T1	T2	T3	T4	T5
HPMC (g)	1	1.15	1.15	1	1
PVP (g)	0.25	-	-	-	-
Methanol (ml)	10	10	15	15	15
PEG 200 (ml)	0.9	0.2	-	-	-
PEG 4000(mg)	-	-	0.25	0.25	0.25
PG (ml)	0.2	0.4	04	0.4	0.4
Glycerine	-	-	-	1 drop	2 drops

(b) Image of the optimized transdermal patch



(c) Characterization of transdermal patches (n=3)

Evaluation	Nicotine	Aspirin	Paracetamol
Folding Endurance	265.34 ± 2.35	265.67 ± 1.67	265.34 ± 1.69
Weight Variation	23.84 ± 0.71 mg	23.97 ± 0.57 mg	24.94 ± 0.2 mg
Percentage Moisture Loss	0.02% ± 0.01	0.02% ± 0.01	0.02% ± 0.02
Percentage Moisture Absorption	0.02% ± 0.01	0.02% ± 0.01	0.02% ± 0.04
Swelling Index	0.67% ± 0.05	0.68% ± 0.02	0.7% ± 0.02
Drug Content	0.15 ± 0.01 µg	0.41 ± 0.04 mg	0.3 ± 0.01 mg

Figure 4: Development of transdermal patch, (a) Composition of different formulations of patch towards its optimization, (b) Image of an optimized transdermal patch, (c) Characterization of nicotine, aspirin, and paracetamol transdermal patch respectively

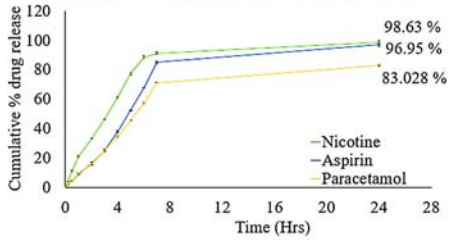
The drug release profile shows that the fast disintegrating sublingual patch is releasing the paracetamol and nicotine faster than aspirin. This is mainly related to the solubility of the drug molecules in water. It also indirectly shows that the drug crystals are heterogeneously dispersed as a drug reservoir in the patch. The drug release profile is further subjected to kinetic analysis and model fitting to (i) zero-order, (ii) first order, (iii) Higuchi model, and (iv) Korsmeyer- Peppas models respectively (figure 3c i-iv). It shows the regression coefficients (R^2) and slope of the curve (n)(for KP) of the graphs. The kinetic analysis and modeling show that the matrix follows non-fickian diffusion with case II transport. Indicating enhanced release at zero-order may be due to the faster disintegration of the sublingual patch. Given the details of the sublingual patch, the transdermal patch is said to analyze.

Transdermal patch

The transdermal patch with the required properties is developed (figure 4). To optimize the transdermal patch with required properties, five formulations were developed (T1 to T5) (figure 4a). The polymer blend of HPMC and PVP is taken to provide a dissolution followed by gelling matrix. However, the blend is phase separating into different phases. Thus, PVP is removed from the blend. For optimizing the plasticizer properties, three different plasticizers is taken initially. Among that PEG 200 is later removed because of the sticky nature of the developed film. A plasticizer combination of PEG 4000 with PG produced required flexibility for the patch. The T5 formulation produced all the required properties of the patch, which is shown in figure 4b.

The optimized transdermal patch is flexible, odorless, and translucent in its physical nature and is loaded with different drugs like nicotine, aspirin, and paracetamol. In figure 4c, the evaluation parameters are shown (weight variation, folding endurance, percentage moisture loss, percentage moisture absorbed, swelling index, and drug content) conducted for the particular drug-loaded patches are given.

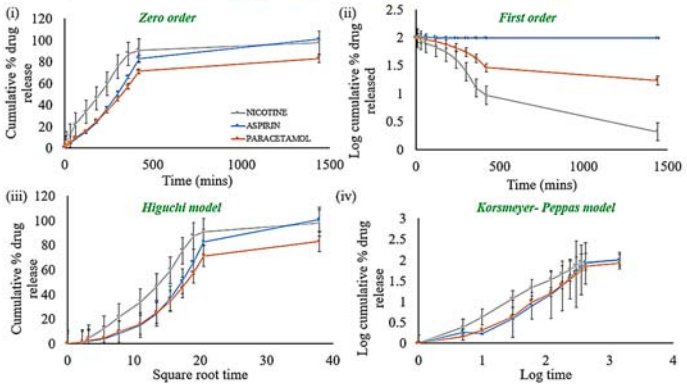
(a) Drug release from transdermal patches



(b) Regression coefficient of transdermal patches

Drug loaded patches	R^2				n
	Zero order	First order	Higuchi model	Korsmeyer-Peppas model	
Nicotine	0.56	0.86	0.84	0.97	0.72
Aspirin	0.7	0.92	0.88	0.94	0.75
Paracetamol	0.73	0.90	0.84	0.94	0.72

(c) Kinetic modelling of drug release from transdermal patches



(d) Combined drug release from sublingual and transdermal patches

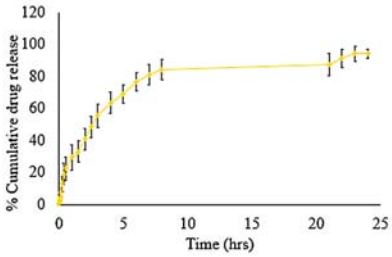


Figure 6: Drug release properties of a transdermal patch, (a) Cumulative % release vs. time profile of nicotine, aspirin, and paracetamol transdermal patch respectively, (b) Regression coefficient of nicotine, aspirin, and paracetamol transdermal patches, (c) Graphs representing kinetic models of nicotine, aspirin, and paracetamol transdermal patches were; (i) Zero-order, (ii) First order, (iii) Higuchi model and (iv) Korsmeyer- Peppas model respectively, (d) Cumulative % drug release vs. time of combination patches

The figure 5a shows the drug release profile of the transdermal drug-loaded patch, releasing 98.63%, 96.95 % and 83.02 % of nicotine, aspirin, and paracetamol respectively in a controlled rate for 6 hrs.

Figure 5b shows the regression values (r^2) obtained from the kinetic and model fitting analysis of release data. The figure 5c shows the graphical representation of the kinetic analysis and model fitting, for various patches (nicotine, aspirin, and paracetamol) for (i) zero-order, (ii) first order, (iii) Higuchi model and (iv) Korsmeyer- Peppas model respectively. It shows a non-fickian transport with first-order kinetics for all the drugs making it a suitable candidate for prolonged-release through transdermal route.

The successfully prepared patches were analyzed together to check the proposed response and the results in Fig.6d shows, in the first 5 mins of administration 19.19% of the drug is released as part of loading dose and carried on to for 95.29% in 24 hrs making the system compatible to use as intended. After analyzing the patches for combined delivery, it is evident that the drug is released in a controlled manner for the prolonged duration by transdermal patch after an initial burst release by sublingual patch. With this effort, we can say the drug-releasing patch is efficient in its properties and can be aimed for various purposes, as mentioned earlier. The therapeutic platform range it covers can be helpful to the widespread population benefiting effectively in multiple advantages.

When combining the three individually equipped systems into a common platform for detecting and treating the nocturnal seizure attacks in neonatal epileptic patients, would turn to be beneficial by allowing the patients to acquire fearless sleep nights. Moreover, detection followed by therapy for prophylaxis is important in several fields like epilepsy and COVID-19. In the case of COVID-19 detection, using RT-PCR can be combined with the delivery of antivirals or NSAIDs using a platform of a sublingual and transdermal patch.

Conclusion

The aim of the work was to develop a model system for "prophynostics" by combining detection and prophylaxis. Here detection of vibration as in seizure followed the delivery of nicotine or NSAIDs through two different routes (sublingual and transdermal) were proposed. Each component was individually analyzed and was found effective individually as well as in combination. Depending on the scenario, this concept of "prophynostics" and the prototype can be explored to treat different diseases such as epilepsy and COVID-19. An addiction-reducing method is beneficial for a large human population when drugs like nicotine are explored for delivery, and for that, this device is helpful. The detection followed by therapy is affordable, cost-effective, and achievable by both urban and rural populations in the country. The future plan is to do detailed studies using this prototype.

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References

1. M. Marta, B. Anna , H.G. Virginia, Precision medicine in variceal bleeding: Are we there yet?, Journal of Hepatology, 72(4), 774-778, (2020)
2. R. Kojima, D. Aubel, M. Fussenegger, Novel theranostic agents for next-generation personalized medicine: small molecules, nanoparticles, and engineered mammalian cells, Current opinion in chemical biology, 28, 29-38, (2015)
3. C. Alvarez-Lorenzo, A. Concheiro, Bioinspired drug delivery systems, Current opinion in biotechnology, 24(6), 1167-73, (2013)
4. X. Chen, S.T. Wong, Cancer theranostics: An introduction, Cancer Theranostics, Elsevier, Academic Press, 3-8 (2014)
5. K.Y. Choi, E.J. Jeon, H.Y. Yoon, B.S. Lee, J.H. Na, K.H. Min, S.Y. Kim, S.J. Myung, S. Lee, X. Chen, I.C. Kwon, Theranostic nanoparticles based on PEGylated hyaluronic acid for the diagnosis, therapy and monitoring of colon cancer, Biomaterials, 33(26), 6186-93, (2012)
6. S.A. Keek, R.T. Leijenaar, A. Jochems, H.C. Woodruff, A review on radiomics and the future of theranostics for patient selection in precision medicine, The British journal of radiology, 91(1091), 20170926, (2018)
7. K. Kamalasanan, Biomimetic Conjoining Pathways for COVID-19 Nanomedicine Drug Discovery and Medical Devices: Prophylactic Medicines as Alternative for Vaccines, Trends Biomater. Artif. Organs, 34(S1), 8-11 (2020)
8. N. Khlebtsov, V Bogatyrev, L Dykman, B Khlebtsov, S Staroverov, A Shirokov, L Matora, V Khanadeev, T Pylaev, N Tsyganova, G Terentyuk, Analytical and Theranostic Applications of Gold Nanoparticles and Multifunctional Nanocomposites, Theranostics, 3(3), 167–180 (2013)
9. Kaladhar K, Zero-dimensional nanotubes as nano-devices to dismantle COVID-19: Possibilities of advanced coatings, "nano-instant vaccines" and "prophynostics", Trends Biomater. Artif. Organs, (accepted) (2020).
10. J. Yang, Y. Zheng Y, X. Gou, K. Pu, Z. Chen, Q. Guo, R. Ji, H. Wang, Y. Wang, Y. Zhou, Prevalence of comorbidities in the novel Wuhan coronavirus (COVID-19) infection: a systematic review and meta-analysis, International Journal of Infectious Diseases, 94, 91-95 (2020)
11. A. Cortegiani, G. Ingoglia, M. Ippolito, A. Giarratano, S. Einav, A systematic review on the efficacy and safety of chloroquine for the treatment of COVID-19, Journal of critical care, 57, 279-283, (2020)
12. A.M. Abbatecola, R. Antonelli-Incalzi, COVID-19 Spiraling of Frailty in Older Italian Patients, The Journal of Nutrition, Health & Aging, 24, 1-3, (2020)
13. Y. Zhang, K. Brown, K. Siebenaler, A. Determan, D. Dohmeier, K. Hansen, Development of lidocaine-coated microneedle product for rapid, safe, and prolonged local analgesic action, Pharmaceutical research, 29(1), 170-177, (2012)
14. C.P. Panayiotopoulos, The Epilepsies: Seizures, Syndromes and Management, Neonatal Seizure and Neonatal Syndromes, Oxfordshire, Bladon Medical Publishing, (2005)
15. A. Becchetti, P. Aracri, S. Meneghini, S. Brusco, A. Amadeo, The role of nicotinic acetylcholine receptors in autosomal dominant nocturnal frontal lobe epilepsy, Frontiers in Physiology 6, 22, 1-12, (2015)
16. I.E. Scheffer, K.P. Bhatia, I. Lopes-Cendes, D.R. Fish, C.D. Marsden, F. Andermann, E. Andermann, R. Desbiens, F. Cendes, J.I. Manson, Autosomal dominant frontal epilepsy misdiagnosed as sleep disorder, Lancet, 343, 51-517, (1994)
17. O.K. Steinlein, Genetics and Epilepsy, Dialogues in Clinical Neurosciences, 10(1): 29-38, (2008)
18. H.A. Phillips, I.E. Scheffer, K.M. Crossland, K.P. Bhatia KP, D.R. Fish, C.D. Marsden, S.J.L. Howell, J.B.P. Stephenson, J. Tolmie, G. Plazzi, O. Eeg-Olofsson, Autosomal dominant nocturnal frontal lobe epilepsy: Genetic Heterogeneity and evidence for a second locus at 15q24, The American Journal of Human Genetics, 63(4), 1108-1116, (1998)
19. I.E. Scheffer, K.P. Bhatia, I. Lopes-Cendes, D.R. Fish, C.D. Marsden, F. Andermann, E. Andermann, R. Desbiens, F. Cendes, J.I. Manson, Autosomal dominant nocturnal frontal lobe epilepsy: a distinctive clinical disorder, Brain, 118, 61–73, (1995)
20. O.K. Steinlein, J.C. Mulley, P. Propping, R.H. Wallace, H.A. Phillips, G.R. Sutherland, I.E. Scheffer, S.F. Berkovic, A missense mutation in neuronal nicotinic acetylcholine receptor a4 subunit is associated with autosomal dominant nocturnal frontal lobe epilepsy, Nature Genetics, 11(2), 201-203, (1995)
21. K. Erik, Sleep and Epilepsy: Strange bedfellows No More, Minerva Pneomol, 50(3): 159-176, (2011)
22. C.E. Allenby, K.A. Boylan, C. Lerman, M. Falcone, Precision medicine for tobacco dependence: development and validation of the nicotine metabolite ratio, Journal of neuroimmune pharmacology, 11(3), 471-83, (2016)
23. R. Niaura, K. Poulas, D. Kouretas, A. Barbouni, A. Tsatsakis, K. Farsalinos, A. Vantarakis, J. Le Houezec, Nicotine and SARS-CoV-2: COVID-19 may be a disease of the nicotinic cholonergic system, [Published online ahead of print, 2020 Apr 30], Toxicol Rep, (2020)
24. C.D. Son, F.J. Moss, B.N. Cohen, H.A. Lester, Nicotine normalizes

intracellular subunit stoichiometry of nicotinic receptors carrying mutations linked to autosomal dominant nocturnal frontal lobe epilepsy, Molecular pharmacology, 75(5), 1137-48, (2009)
25. J.M. Oakes, R.M. Fuchs, J.D. Gardner, E. Lazartigues, X. Yue, Nicotine and the renin-angiotensin system, Am J Physiol Regul Integr Comp Physiol, 315(5), R895 R906 (2018)
26. J.P. Changeux, Z. Amoura, F.A. Rey, M. Miyara, A nicotinic hypothesis for Covid-19 with preventive and therapeutic implications, Reports, Biology, 343(1), 33-39, (2020)
27. J.M. Leung, C.X. Yang, D.D Sin, COVID-19 and nicotine as a Mediator of ACE-2, European Respiratory Journal, 55(6), 2001261, (2020)
28. P. Little, Non-Steroidal anti-inflammatory drugs and covid-19, BMJ, 368, m1185, (2020)
29. J.O. Willoughby, K.J. Pope, V. Eaton, Nicotine as an antiepileptic agent in ADNFLE: an N-of-one study, Epilepsia, 44(9), 1238 1240, (2003)
30. A.J. Scott, J.R. Mabesa Jr, C. Hughes, D.J. Gorsich, G.W. Auner, Equipping small robotic platforms with highly sensitive more accurate nuclear, biological, and chemical (NBC) detection systems, In Unmanned Ground Vehicle Technology V, 5083, 539-548, (2003)
31. X. Zhang, S. Fronz, N. Navab, Visual marker detection and decoding in ar systems: A comparative study, In Proceedings, International Symposium on Mixed and Augmented Reality, 97-106, (2002)
32. N. Arunkumar, V.S. Balaji, S. Ramesh, S. Natarajan, Automatic detection of epileptic seizures using independent component analysis algorithm, In IEEE -International Conference On Advances In Engineering, Science And Management, 542-544, (2012)
33. U.R. Acharya,F. Molinari, S.V. Sree, S. Chattopadhyay, K.H. Ng, J.S. Suri, Automated diagnosis of epileptic EEG using entropies, Biomedical Signal Processing and Control, 7(4), 401-8, (2012)
34. A.S. Kumar, T. Kuruvila, K. SreeGanga, R. Radhakrishnan, A.J. Nair, K. Kamalasanan, Early Detection of Seizures in Epilepsy Using Point Of Care (POC) Systems, Journal of Pharmaceutical Sciences and Research, 9(3), 302-306, (2017)
35. S. Amano, M. Yokoyama, R. Torii, J. Fukuoka, K. Tanaka, N. Ihara, F. Hazama, High performance seizure – monitoring system using a vibration sensor and videotape recording: Behavioral analysis of genetically epileptic rats, Laboratory Animal Science, 47(3), 317-320 (1997)
36. A. Shoeb, H. Edwards, J. Connolly, B. Bourgeois, S.T. Treves, J. Guttag, Patient-specific seizure onset detection, Epilepsy & Behavior, 5(4), 483-98, (2004)
37. H. Motoe, S. Koide, Cradle device and electronic apparatus connectable to cradle device, Toshiba Corp, United States patent application, US 12/335,415, (2009)
38. A. Ulate-Campos, F. Coughlin, M. Gaínza-Lein, I.S. Fernández, P.L. Pearl, T. Loddenkemper, Automated seizure detection systems and their effectiveness for each type of seizure, Seizure, 40, 88-101, (2016)
39. Y. Bai, L. Yao , T. Wei, F. Tian, D.Y. Jin, L. Chen, M. Wang, Presumed asymptomatic carrier transmission of COVID-19, Jama, 323(14), 1406-7, (2020).
40. T. Sanner, T.K. Grimsrud, Nicotine: Carcinogenicity and Effects on Response to Cancer Treatment - A Review, Front Oncol, 5, 196, (2015)
41. N.L. Benowitz, Nicotine addiction, New England Journal of Medicine, 362(24), 2295-303, (2010)
42. A.B. Khan, T. Kingsley, P. Caroline, Sublingual tablets and the benefits of the sublingual route of administration, Journal of Pharmaceutical Research, 16(3), 257-67, (2017)
43. B. Bhyan, S. Jangra, Formulation and evaluation of fast dissolving sublingual films of Rizatriptan Benzoate, International Journal of Drug Development and Research, 4(1), 133-143, (2012)
44. R.P. Mark, L. Robert, Transdermal drug delivery, Nature Biotechnology, 26(11), 1261-1268, (2008)
45. R. Soltero, R. Krailler, J. Czeisler, The effects of pH, ionic concentration and ionic species of dissolution media on the release rates of quinidine gluconate sustained release dosage forms, Drug development and industrial pharmacy, 17(1), 113-40, (1991)
46. V.V Prashant, Y. Mamatha, S. Arunkumar, S. Mathew, A. Abraham, Formulation and evaluation of mucoadhesive buccal patches of Aceclofenac, Der Pharmacia Lettre, 4(1), 297-306, (2012)
47. D. D'cruz, A. Ajith, T. Subburaj K. Kamalasanan, Magnesium stearate is an incompatible excipient for aspirin in wet granulation producing non-linear degradation, Journal of Pharmaceutical Sciences and Research., 10(2), 240-242, (2018)
48. S.T. Prajapati, C.G. Patel, C.N. Patel, Formulation and evaluation of transdermal patch of Repaglinide, International Scholarity Research Network, 651909, (2011)
49. S.N.R. Adhikari, B. S. Nayak, A.K. Nayak, B. Mohanty, Formulation and evaluation of buccal patches for delivery of atenolol, American Association of Pharmaceutical Scientists PharmSciTech, 11, 1038-1044, (2010)
50. M. Alagusundaram, B. Chengaiah, S. Ramkanth, S.A. Parameswari, C. Madhu, D. Dhachinamoorthi, Formulation and evaluation of mucoadhesive buccal films of ranitidine, International Journal of Pharmtech Research, 1(3), 557-563, (2009)
51. A.R. Augusthy, K.V. Vipin, S.C. Chandran, A.V. Muhammed Nizar, K. Sreeraj, Formulation and evaluation of rabeprazole buccal patches, Journal of Pharmaceutics and Nanotechnology, 2(3), 23-30, (2014)
52. V. Kumar, F. Zakir, G. Agarwal, A. Choudhary, Formulation and evaluation of buccal patches of venlafaxine, International Journal of Pharmacy and Biological Sciences, 1(3), 170-182, (2011)
53. S. Cherukuri, U.R. Batchu, K. Mandava, V. Cherukuri, K.R. Ganapuram, Formulation and evaluation of transdermal drug delivery of topiramate, International Journal of Pharmaceutical Investigation, 7(1), 10-17, (2017)
54. P. Costa, J.M.S. Lobo, Modeling and comparison of dissolution profiles, European Journal of Pharmaceutical Scineces, 13, 123-133, (2001)
55. M.C. Sreekanth, A. Ajayakumar, K. Santhi, Formulation and evaluation of buccal patches of carvedilol phosphate, International Research Journal of Pharmacy, 7(8), 23-27, (2016)
56. J. Rajeev, S. Mukundan, B.K. Raj, V.P. Dhanaja, T. Subburaj, K. Kamalasanan, Formulation of novel immediate release heterolithic buccal patch of aspirin, Journal of Pharmaceutical Sciences and Research, 10(8), 2079-2083, (2018)